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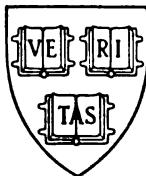
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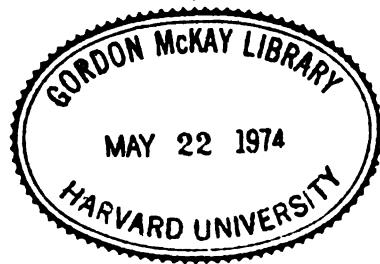
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SOIL BIOLOGY

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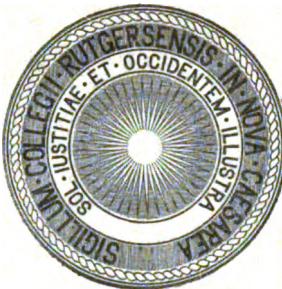
- Page 253, legend for figure 7, "Osmeter" should read "Osmometer."
Page 296, third line above footnote, "exacctly" should read "exactly."
Page 299, table 3, part 1, Average for sample A, "0.038" should read "0.030."
Page 307, table 10, title, "alum num" should read "aluminum."
Page 307, four lines at foot of page should be inserted between ninth and tenth lines from
foot of page 308.
Page 413, table 25, first line of figures, "0" should read "3."
Page 420, table 31, second line of figures, "2.77" should read "6.77."

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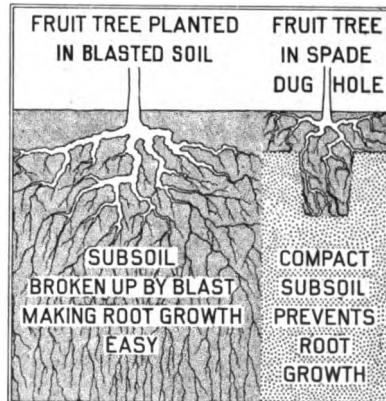
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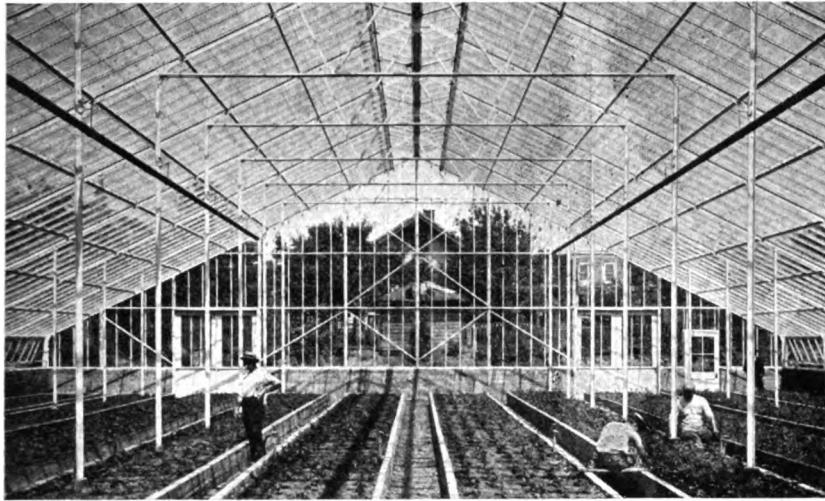
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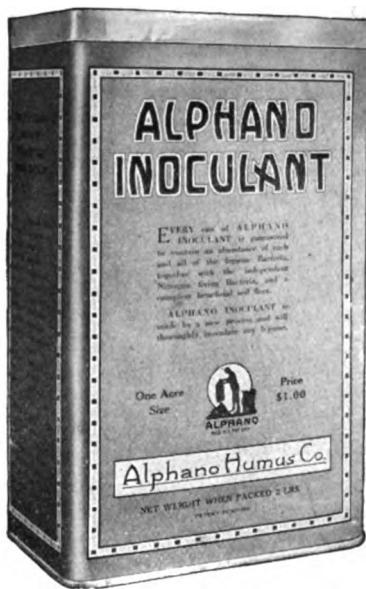
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THE RELATION OF GREEN MANURES TO NITROGEN FIXATION¹

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INTRODUCTION

The lack of nitrogen and organic matter in many cultivated soils is noted by a decrease in the crop yield. This deficiency is most apparent in the older farmed soils where cultivation and removal of crops has been practiced for a number of years. In order that crop production may be increased, it is important to study the most economical method for supplying nitrogen and organic matter. The turning under of green manures offers a possible solution. Aside from the plant-food returned with the green manure, it frequently benefits the physical and biological properties of the soil. The total number of soil microorganisms, as well as the different groups of microorganisms, is markedly influenced by the addition of green manure. This effect on the soil flora is reflected in the fertilizing constituents made available for plant growth.

Many investigations have been directed toward this phase of the problem and much valuable information has been obtained concerning the relation of green manures to biologic processes in soil. From a review of the literature, it appears that the effect of green manures on nitrogen fixation deserves further study. Accordingly, experiments were planned to investigate the relation of green plant tissue to free nitrogen-fixing organisms. The nature of the problem may be seen from the following brief review of the literature.

HISTORICAL

Berthelot (2) was the first to note a gain of nitrogen through the action of biological agents on the humus of the soil. In 1902 Henry (9) observed an increase in nitrogen when forest leaves were allowed to decompose in soil. His results were confirmed by Sückting (25) and Hornberger (11).

Koch (15) found a 10 per cent increase in total nitrogen of soil which had been allowed to stand for $2\frac{1}{2}$ years. This increase he attributed to the assimilation of nitrogen by the non-symbiotic nitrogen-fixing organisms. Krzemieniewski (18) studied the gain in nitrogen with pure cultures of *Azotobacter chroococcum*, *Radiobacter*, and *Granulobacter* in solutions containing soil

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humate. All cultures showed an increase in nitrogen from the use of humate, especially when the first two organisms were grown together. Remy and Rösing (23) obtained similar results with the use of soil humate. They concluded that the iron contained in the humate probably acted as a stimulant for the nitrogen-fixing organisms.

In a sandy soil, Koch and his associates (16) obtained nitrogen fixation by the addition of dextrose, cane sugar, or starch. There was practically no increase shown when straw, filter paper, or buckwheat was applied. The addition of green mustard in a dried, pulverized form resulted in a loss of nitrogen, which loss, they thought, was due partly to the escape of ammonia from the decomposing mustard.

Koch (13) showed that the beneficial effect of sugar, i.e., gain in nitrogen, may be noted by an increase in crop yield. During the first year no increase was noted; however, in the second and third years the treated series showed an increase in crop yield. In a later publication (14) the same author noted that cellulose may be used as a source of energy for nitrogen-fixing organisms provided the time of incubation is long enough for the cellulose to decompose. The greatest gain in nitrogen occurred where the largest amount of cellulose was digested. For each gram of carbohydrate consumed, cellulose gave a greater fixation than dextrose.

According to Hutchinson (12) winter applications of sugar and starch benefited the crop yield. However, spring applications failed to give an increase. He attributed this difference to the effect of the season on *Azotobacter*. Hoffmann (10) found an increase of nearly 1000 pounds of nitrogen per acre as a result of three years' treatment of soil with sugar. Similar results were obtained with starch, but the gain was not so marked. In 1910, Marr (20) reported that straw alone when added to soil increased nitrogen fixation; sugar, also, favored this process.

Experiments carried out by Pringsheim (22) showed that the nitrogen-assimilating organisms can make use of decomposing cellulose as a source of energy. Recent investigations by Bottomley (3) have shown that soluble humates may be used by the nitrogen-assimilating organisms.

Hanzawa (8) employed humus from stable manure and from green manure as energy sources for *Azotobacter*. In solution he noted that stable-manure humus increased nitrogen fixation, while green-manure humus proved injurious. However, only a very limited number of determinations were made with green manures.

Löhnis and Green (19) carried on similar experiments. They worked with mixed cultures of *Azotobacter chroococcum*, *Beijerinckii*, *Vinelandii*, and *Vitreum*. Beijerinck's mannit solution² was used with the addition of about 0.2 gm. of dried material to each 100 cc. portion of liquid. They compared the gain in nitrogen with stable manure, green (pea) manure, peat, and straw. Their results are given in the following table:

² Tap water, 1,000 cc.; mannit, 20.0 grams; dipotassium phosphate, 0.5 grams.

MATERIAL	NITROGEN ASSIMILATED IN 100 CC. OF SOLUTION AFTER THREE WEEKS
	mgm.
Fresh straw.....	10.0
Fresh stable manure.....	9.8
Fresh peat.....	9.3
Green manure.....	8.0
Beijerinck's mannit solution alone.....	5.6

The results show that all of the organic substances, when added to the mannit solution, aided nitrogen fixation. Apparently straw, stable manure, and peat furnished the best sources of energy for the nitrogen-fixing organisms. These investigators stated that more nitrogen was assimilated by the mixed cultures than by any one of the individual strains.

When the stable manure, green manure, peat, and straw were converted into humus under aerobic or anaerobic conditions, the effect on nitrogen fixation was as follows:

MATERIAL	CONDITIONS FOR HUMUS FORMATION	NITROGEN ASSIMI- LATED IN 100 CC. OF SOLUTION AFTER THREE WEEKS	
		mgm.	mgm.
Stable manure.....	Aerobic	14.4	
	Anaerobic	13.6	
Green manure.....	Aerobic	9.0	
	Anaerobic	8.7	
Peat.....	Aerobic	9.9	
	Anaerobic	10.9	
Straw.....	Aerobic	10.4	
	Anaerobic		

After humification, the stable manure takes first place as an energy source for the free nitrogen-fixing organisms. This difference may be due to the higher content of cellulose material. Here, unlike Hanzawa's results, green-manure humus, as well as the green manure, enhanced nitrogen fixation.

In his study of the carbon and nitrogen balance in the soil, Felber (7) applied dried, pulverized straw and obtained nitrogen fixation in both sand and clay soils. Brown and Allison (4) treated soil in pots with stable manure, straw, stover, and hay. Both leguminous and non-leguminous hays were employed. A gain in nitrogen was the result of these treatments. The tissue from non-legumes gave a greater gain than that from legumes.

Because of their bearing on the experiments described in this paper, the results of Dvorak (6) deserve special consideration. He found that varying amounts of nitrogen are fixed when *Azotobacter* is grown in a solution containing carbohydrates from different sources. Comparisons were made of the amount of nitrogen fixed in the presence of glucose with that fixed from

oak leaves, clover, maple leaves, straw, alfalfa, corn stalks, lupine, corn roots and stubble, and pine needles. These materials were dried, ground, and 10-gram portions added to 250 cc. of tap water containing 1 per cent of calcium carbonate and 1 per cent of dibasic potassium phosphate, except clover, lupine, and alfalfa which were added in a green, crushed form. This solution was placed in 2-liter Erlenmeyer flasks, sterilized, and inoculated with a pure culture of *Azotobacter chroococcum*. The gain in nitrogen is shown in the following table adapted from Dvorak's results.

MATERIAL	NITROGEN ASSIMILATED IN 100 CC. OF SOLUTION AFTER FOUR WEEKS
	mgms.
Glucose.....	23.04
Corn roots and stubble.....	7.36
Clover.....	7.12
Straw (wheat).....	5.60
Corn stalks.....	4.48
Oak leaves.....	2.24
Lupine.....	2.20
Alfalfa.....	1.72
Maple leaves.....	1.48
Pine needles.....	0.88

The value of corn stalks for nitrogen fixation is clearly shown by the figures in the table. Of all the substances, with the exception of glucose, corn stalks gave the highest gain in nitrogen. In a descending scale, clover and wheat straw rank next in stimulating nitrogen fixation. The indirect beneficial effect of plant tissue on nitrogen fixation is clearly established from the results given in this table. Dvorak concluded that the greatest gain in atmospheric nitrogen by *Azotobacter chroococcum* was obtained from compounds with low carbon and with high oxygen content. He thought the small fixation of nitrogen in the case of pine needles was due partly to the presence of terpenes and tannic acid.

Stoklasa (24) tested the availability of various sugars for *Azotobacter chroococcum*. A comparison of arabinose, saccharose, xylose, glucose, galactose, fructose, rhamnose, lactose, and maltose showed that in liquid cultures the highest fixation of nitrogen occurred with arabinose and the lowest with rhamnose.

When *Azotobacter* was grown on polysaccharides, various alcohols, sugars, aliphatic and non-aliphatic organic acids, substances occurring widely in nature Mockeridge (21) noted fixation of nitrogen by this organism.

From the preceding data, it seems certain that in a soil with the proper reaction, the common soil flora should give an increase in nitrogen provided a suitable source of energy is added. Based on these results, a series of experiments was planned to study the effect of green manures on nitrogen fixation.

METHODS

Two common field crops, a legume and a non-legume, were selected. These plants were chosen because of their difference in content of nitrogen and pentosan. The results of many analyses by Düring (5) show that non-legumes are, as a rule, much higher in pentosans than are legumes. Since pentosans are easily hydrolyzed to pentose sugars, they are important from the standpoint of the independent nitrogen-fixing organisms.

Red clover (*Trifolium pratense*), oats (*Avena sativa*), and wheat (*Triticum vulgare*) were used as green manures. The plants were cut into fine pieces with scissors and 1.5 per cent of the green tissue added to the soil, or 0.5 per cent on the dry basis. This amount was selected because it approximates a good crop of wheat, oats, or clover. Total nitrogen analyses were made of five representative samples of tissue at the time it was added to the soil. The average of these analyses is given in the tabular data under the heading "nitrogen added to 100 grams of soil." The nitrogen of the green manure was subtracted from the total analyses of the treated soil.

Two types of soil were employed, one a field soil classed as Miami silt loam, and the other a garden soil classed as a member of the Clyde series. These two soils were selected because of their difference in organic matter. The field soil is medium low, while the garden soil is high in organic substances. The soils were sieved thoroughly, mixed, and 400-gram portions on the dry basis were weighed out. The green tissue was then added to the soil and the whole thoroughly mixed. In order to secure aeration, the soil was placed in shallow soup plates. These were incubated at 25°C. The soil moisture was maintained at 25 per cent, which represented about half saturation. At regular intervals, the loss by evaporation was replaced by the addition of distilled water.

The first total nitrogen determination was made after 2 weeks and again after 4, 6, and 8 weeks. For each analysis one plate of soil from each similarly treated group was used. The soil was dried, pulverized, and thoroughly mixed on glazed paper. Ten-gram samples were then taken for analysis.

In all nitrogen determinations the greatest care was exercised. Only the purest chemicals were used. The contents of the flasks were digested until clear and the heating continued for another hour to insure complete digestion. All analyses, unless otherwise stated, were made according to the Kjeldahl method modified to include nitrate nitrogen. The reagents were made up in amounts large enough to last throughout the work, thus insuring greater uniformity in results. The standard acid and alkali were frequently checked from time to time. The period of distillation lasted for at least an hour. The standard acid and alkali in these experiments were of N/10 normality. Cochineal was used as an indicator.

Nitrogen-fixation in garden soil treated with green clover and green wheat -

A series of plates was arranged as follows:

- a. Four plates of garden soil untreated.
- b. Four plates of garden soil plus 1.5 per cent of green clover.
- c. Four plates of garden soil plus 1.5 per cent of green wheat.

The results of this experiment are recorded in table 1.

TABLE 1
Effect of green clover and green wheat on nitrogen fixation in garden soil

NO.	TREATMENT	NITROGEN ADDED TO 100 GRAMS OF SOIL	NITROGEN IN 100 GM. OF SOIL											
			After two weeks			After four weeks			After six weeks			After eight weeks		
			Total	Average	Increase	Total	Average	Increase	Total	Average	Increase	Total	Average	Increase
1	None	None	413.0			410.2			410.2			406.0		
2	None	None	417.2			410.2			411.6			407.4		
3	None	None	417.2			409.8			407.4			406.0		
4	None	None	417.2			411.6			410.2			403.2		
5	None	None	417.2			407.4			407.4			407.4		
6	1.5 clover	13.2	423.6*			413.8			413.8			406.8		
7	1.5 clover	13.2	419.4			412.4			413.8			405.4		
8	1.5 clover	13.2	422.2			411.0	412.7	2.9	412.4	412.7	3.3	408.2		
9	1.5 clover	13.2	418.0			413.8			412.4			408.2		
10	1.5 clover	13.2	423.6			412.4			411.0			411.0		
11	1.5 wheat	11.5	425.3			412.7			415.5			416.9		
12	1.5 wheat	11.5	426.7						415.5			411.3		
13	1.5 wheat	11.5	418.3			409.9	412.3	2.5	414.1	413.8	4.4	411.6		
14	1.5 wheat	11.5	416.9			414.1			411.3			414.1		
15	1.5 wheat	11.5	423.9			412.7			412.7			404.3		

* The green manure nitrogen was subtracted from the total nitrogen of the treated soil.

Different plates of soil were used for each analysis instead of taking samples of soil from the same plate. The decrease in nitrogen of the untreated soil after four, six, and eight weeks is due most probably to a change in methods and not to any actual loss of nitrogen. All the figures given in the vertical columns represent the results of analyses made at the same time, i.e., the methods followed were alike.

It will be seen by an examination of the vertical columns of table 1 and figure 1 that in every case the treated soil, after allowance is made for the green-manure nitrogen present, showed a gain in nitrogen over the untreated. There was some variation in the amount of nitrogen assimilated depending

upon the period of incubation. The greatest gain was obtained two weeks after the green manure was added.

A comparison of these two green manures indicates that clover is more beneficial during the early stages of decomposition, while wheat is beneficial for a much longer period of time. Apparently the non-legume gives a greater gain in nitrogen than the legume. This difference in favor of wheat is perhaps due to the higher content of carbohydrate material.

Under the conditions of this experiment, it seems safe to conclude that green manures as clover and wheat when added to soil furnish some substance which benefits the nitrogen-fixing flora.

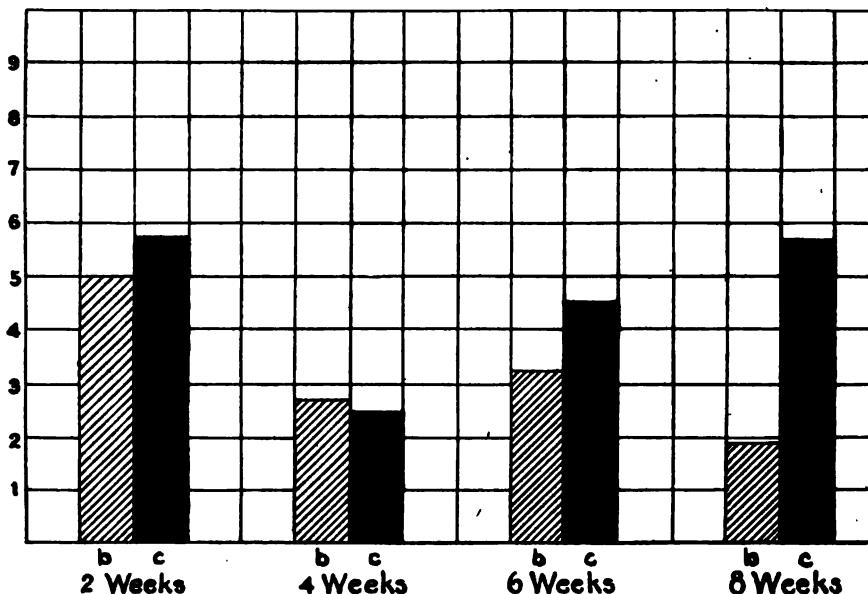


FIG. 1. MILLIGRAMS OF NITROGEN FIXED IN 100 GRAMS OF GARDEN SOIL AS A RESULT OF ADDING GREEN MANURE

b, clover-treated soil; c wheat-treated soil.

Nitrogen fixation in field soil treated with green clover and with green oats

In order to measure the effect of green manures on nitrogen fixation in field soil, a series of plates was arranged similar to those in the preceding test, with the exceptions that green wheat was replaced by green oats and a different sample of green clover was used. The results of previous study showed that the field soil was lower than the garden soil in both organic matter and the total number of bacteria. For these reasons it seemed desirable to make a study of the effect of green manuring on nitrogen fixation in field soil. The arrangement of the experiment and the results secured are shown in table 2.

From the data in table 2 and figure 2, it will be seen that in every case with field soil the treated soil showed a gain in nitrogen beyond that of the untreated. The results, in many instances, are similar to those of the preceding test. The greatest gain of nitrogen was shown two weeks after the green manures were added. The amount of nitrogen fixed varied at the different incubation periods.

Here, as in table 1, a comparison of these two green manures indicates that clover is more beneficial during the early stages of decomposition, while oats are beneficial for a much longer period of time. From the data obtained with

TABLE 2
Effect of green clover and green oats on nitrogen fixation in field soil

NO.	TREATMENT	NITROGEN ADDED TO 100 GMS. OF SOIL	NITROGEN IN 100 GRAMS OF SOIL											
			After two weeks			After four weeks			After six weeks			After eight weeks		
			Total	Average	Increase	Total	Average	Increase	Total	Average	Increase	Total	Average	Increase
	per cent	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
1	None	None	274.4			273.0			277.2			284.2		
2	None	None	274.4			274.4			280.0			285.6		
3	None	None	271.6	272.4		275.8	275.2		277.2	279.7		285.6		
4	None	None	270.2			277.2			281.4			287.0		
5	None	None	271.6			275.8			282.8			285.6		
6	1.5 clover	15.3	281.5			280.1			284.3			288.5		
7	1.5 clover	15.3	281.5			281.5			281.5			288.5		
8	1.5 clover	15.3	285.7	283.2	10.8	281.5	280.1	4.9	280.1	282.1	2.4	287.1	289.1	3.5
9	1.5 clover	15.3	281.5			278.7			280.1			289.9		
10	1.5 clover	15.3	285.7			278.7			284.3			291.3		
11	1.5 oats	7.3	282.5			282.5			282.5			292.3		
12	1.5 oats	7.3	283.9			282.5			288.1			292.3		
13	1.5 oats	7.3	279.7	281.9	9.5	283.9	284.2	9.0	282.5	284.5	3.8	290.9	292.2	6.6
14	1.5 oats	7.3	283.9			282.5			285.3			292.3		
15	1.5 oats	7.3	279.7			289.5			283.9			293.1		

field soil, it is apparent that the non-legume gives a greater gain in nitrogen than the legume. This difference is most likely due to a higher content of carbohydrate material in the oats than in the clover.

There is considerable variation in nitrogen in soil receiving the same treatment when examined at different intervals. These samples, as in the preceding experiment, were taken from different plates throughout the test instead of from the same plate. However, in every analysis the treated soil showed a gain in nitrogen beyond that of the untreated when examined at the same time. In comparing the figures of table 1 with those of table 2 the results

indicate that field soil responds to green manuring, i.e., nitrogen fixation, better than the garden soil.

It appears that green manures as clover and oats when added to soil furnish some substance which benefits the nitrogen-fixing flora.

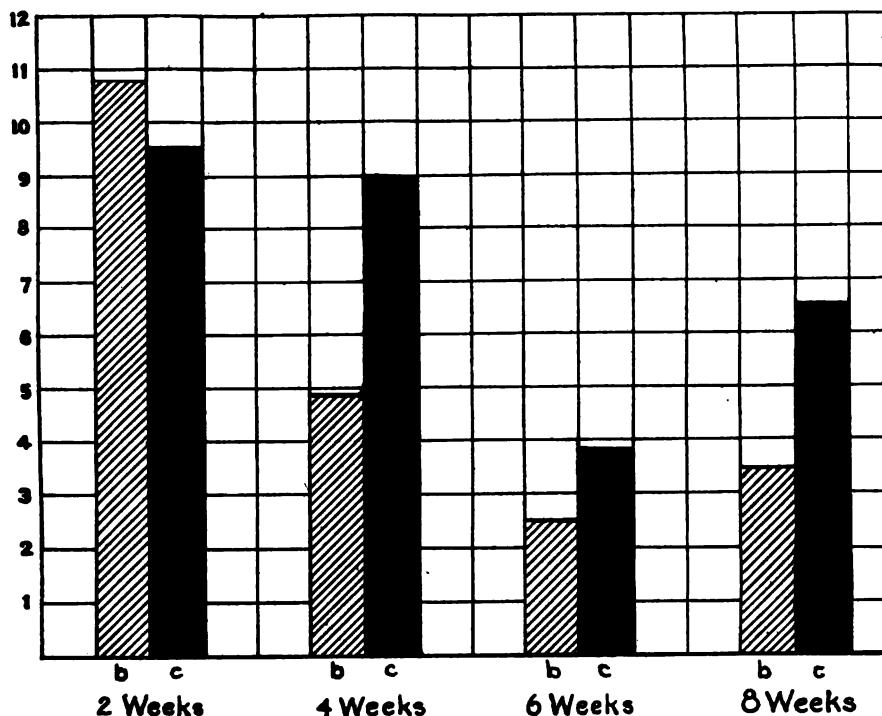


FIG. 2. MILLIGRAMS OF NITROGEN FIXED IN 100 GRAMS OF FIELD SOIL AS A RESULT OF ADDING GREEN MANURE

b, clover-treated soil; c, oats-treated soil.

Nitrogen fixation in field soil treated with green clover, green oats, and 0.5 per cent of mannit

This experiment was made in order to determine whether an increase in nitrogen could be shown with the addition of a green manure in the presence of a small amount of mannit. The treatment was as follows:

- Two plates of field soil plus 0.5 per cent of mannit.
- Two plates of field soil plus 0.5 per cent of mannit plus 1.5 per cent of green clover.
- Two plates of field soil plus 0.5 per cent of mannit plus 1.5 per cent of green oats.

The samples of this series were analyzed after four and eight weeks. The results of this work are presented in table 3.

In reviewing the data in table 3 and figure 3, it will be clearly seen that soil treated with green manure and mannit gave a decided gain in nitrogen beyond that obtained with the mannit alone. Here, as in the preceding experiments, the amount of nitrogen fixed varied with the time of incubation, especially with oats. The greatest gain is noted with oats 4 weeks after treatment. While the results vary somewhat, there is shown in every case a beneficial effect from adding green manures to soil of which the nitrogen-fixing flora had previously received a stimulus by the addition of mannit.

TABLE 3
Effect of green clover and green oats on nitrogen fixation in field soil containing 0.5 per cent of mannit

NO.	TREATMENT	NITROGEN ADDED TO 100 GRAMS OF SOIL <i>per cent</i>	NITROGEN IN 100 GRAMS OF SOIL					
			After four weeks			After eight weeks		
			Total	Average	Increase	Total	Average	Increase
1	None	None	282.8			289.8		
2	None	None	275.8			284.2		
3	None	None	277.2	276.9		289.8	286.2	
4	None	None	274.4			282.8		
5	None	None	274.4			284.2		
6	1.5 clover	15.3	277.3			291.3		
7	1.5 clover	15.3	278.7			289.9		
8	1.5 clover	15.3	281.5	279.2	2.3	291.3	290.2	4.0
9	1.5 clover	15.3	280.1			287.1		
10	1.5 clover	15.3	278.7			291.3		
11	1.5 oats	7.3	290.9			289.5		
12	1.5 oats	7.3	293.7			289.5		
13	1.5 oats	7.3	292.3	292.3	15.4	290.9	289.8	4.0
14	1.5 oats	7.3	295.1			290.9		
15	1.5 oats	7.3	289.5			288.1		

Therefore, under the conditions of this experiment, it seems safe to conclude that the green clover and the green oats benefited the nitrogen-fixing organisms.

Effect of green manures on nitrogen fixation in solution

If green manures cause an increase in the number of nitrogen-fixing bacteria it seems probable that green-manured soil will give a corresponding gain when used to inoculate Ashby's (1) mannit solution. Aside from the greater number of nitrogen-fixing bacteria, it is very probable that the mannit added in the inoculum will have a beneficial effect on nitrogen fixation.



FIG. 3. MILLIGRAMS OF NITROGEN FIXED IN 100 GRAMS OF FIELD SOIL AS A RESULT OF ADDING GREEN MANURE TO SOIL ADDITIONALLY TREATED WITH MANNIT

a, soil with 0.5 per cent mannit; *b*, clover-treated soil + 0.5 per cent mannit; *c*, oats-treated soil + 0.5 per cent mannit.

In order to measure the effect of green manuring on the nitrogen gain in solution, 500 cc. Erlenmeyer flasks, each containing 100 cc. of mannit solution, were inoculated with 10 gram portions of soil. Six flasks were inoculated from each plate of soil. One-half of these were immediately sterilized to serve as controls. These inoculations were made at the beginning of the experiment and repeated at intervals of two weeks, i.e., zero, two, four, six, and eight-

TABLE 4
Effect of green clover and green wheat on nitrogen fixation of garden soil in solution

NO.	TREATMENT	GREEN MANURE NITROGEN ADDED IN 100 CC. OF SOLUTION	NITROGEN IN 100 CC. OF SOLUTION AFTER									
			No weeks Incubated 16 days		Two weeks Incubated 10 days		Four weeks Incubated 7 days		Six weeks Incubated 14 days		Eight weeks Incubated 14 days	
			Total	Increase due to green manure	Total	Increase due to green manure	Total	Increase due to green manure	Total	Increase due to green manure	Total	Increase due to green manure
<i>per cent</i>												
1	None sterile	None	41.02		41.58		44.38		41.72		42.98	
2	None sterile	None			41.44		43.40		42.14		42.88	
3	None sterile	None	41.02		41.72		43.40		42.56		43.40	
4	None not sterile	None	49.42		50.26		53.06		51.80		50.96	
5	None not sterile	None	49.28		50.96		52.92		52.36		50.40	
6	None not sterile	None	49.70		51.52		52.08		50.96		52.08	
7	1.5 clover sterile	1.32	42.08		41.66		42.78		41.94		42.22	
8	1.5 clover sterile	1.32	41.80		42.36		42.92		41.52			
9	1.5 clover sterile	1.32	42.64		42.22		42.54		41.94		43.34	
10	1.5 clover not sterile	1.32	51.88		51.88		53.00		52.58		52.86	
11	1.5 clover not sterile	1.32		1.27	51.88	0.60	52.72	0.92		1.88	53.56	1.73
12	1.5 clover not sterile	1.32	51.88		52.28		52.16		51.88		51.88	
13	1.5 wheat sterile	1.15	41.55		41.13		42.39		41.97		41.83	
14	1.5 wheat sterile	1.15	40.71		41.69				41.69		41.83	
15	1.5 wheat sterile	1.15	41.41		41.55		42.39		41.83		41.55	
16	1.5 wheat not sterile	1.15	50.65		55.85		53.31		50.79		50.93	
17	1.5 wheat not sterile	1.15	50.23	1.13	54.15	3.78	52.61	1.49	51.35	0.69	51.77	1.27
18	1.5 wheat not sterile	1.15	51.49		54.71		52.61		51.07		50.93	

week periods after the manure was added. All of the cultures were kept at 25°C. The time of incubation varied from seven to sixteen days; for the first set of inoculations, sixteen days; for the second set, ten days; for the third, seven days; and for the fourth and fifth tests, fourteen days. It was observed by Koch and Seydel (17) that *Azotobacter* assimilated the greater

part of its nitrogen in the early stages of growth. In order to determine whether the organisms assimilating nitrogen were stimulated soon after inoculations were made, the cultures were analyzed after short periods of incubation. The maximum length of incubation did not exceed sixteen days.

At the end of each incubation period a total nitrogen determination of each flask with the Kjeldahl method not to include nitrates, was made. The results of the analyses are recorded in table 4.

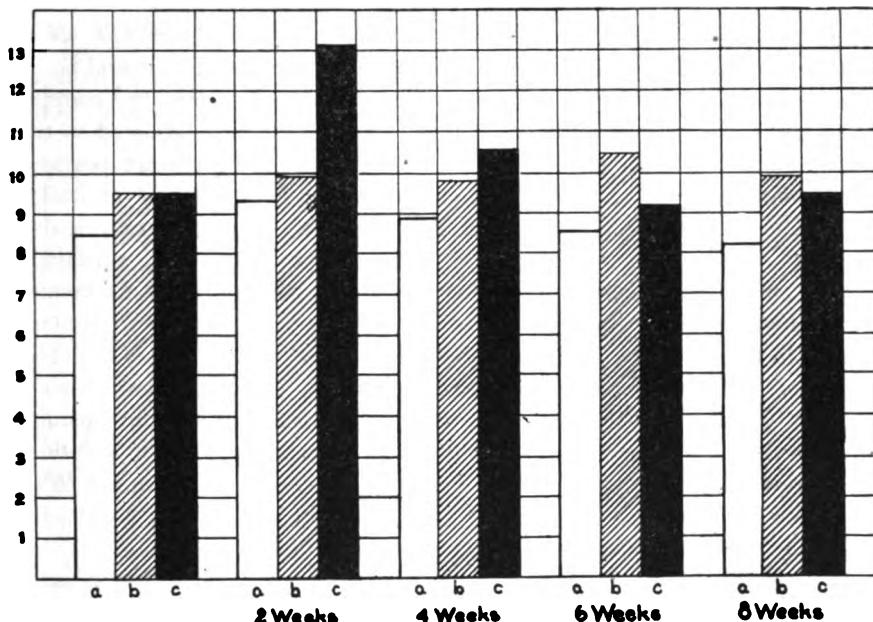


FIG. 4. MILLIGRAMS OF NITROGEN FIXED IN 100 CC. OF ASHBY'S SOLUTION INOCULATED WITH TREATED AND UNTREATED SOIL

Each set was inoculated at intervals of two weeks after starting the experiment, and these allowed to incubate for varying periods. *a*, untreated soil; *b*, clover-treated soil; *c*, wheat-treated soil.

The data in table 4 and figure 4 show that in every case green manures enhanced nitrogen fixation. Although the gain in nitrogen is not large, however, because of a number of parallel determinations, it seems safe to conclude that these substances benefit the fixation of nitrogen in solution. Wheat did not prove to be superior to clover as a stimulus to nitrogen fixation in solution. Clover gave the best results from inoculations made at the beginning of the treatment, and after six and eight week periods. Wheat did better than clover after decomposing for two and four weeks. However, the maximum gain was noted with wheat after two weeks. In general these results fail to show any relation between the age of decomposing manure and the amount of nitrogen assimilated.

This increase in fixation may be due to one or more agencies: first a greater number of nitrogen-fixing bacteria; and second, to the green manure added in the inoculum. Because of the small amount of green manure in the inoculum, it seems that the greater number of bacteria, as a result of the soil treatment, is probably the chief factor concerned with the gain in nitrogen.

Fixation of nitrogen by pure cultures of bacteria

For this test green wheat (*Triticum vulgare*) and red clover (*Trifolium pratense*) were crushed into a semi-fluid mass by grinding for several hours in a porcelain ball mill. To 1 liter of Ashby's solution, minus the mannit, 50 grams of the crushed tissue were added and the reaction adjusted to the neutral point of phenolphthalein. One hundred cubic-centimeter portions of the mixture were measured and poured into 1-liter Erlenmeyer flasks. Each 100 cc. portion represented 5 grams of green tissue or 1 gram of dry material. Both the oats and clover were near maturity and contained smaller amounts of moisture than the preceding samples. The flasks and contents were then sterilized in the autoclave and inoculated with a pure culture of *Azotobacter chroococcum* which had been grown on a medium of the same formula. This same culture of *Azotobacter* had in previous tests shown a gain of 7.4 mgm. of nitrogen when grown in a 1 per cent mannit medium. One-half of the culture flasks were sterilized immediately after inoculation. These were used as controls. The entire set of flasks was placed in the 26°C. incubator for 28 days. At the end of this period total nitrogen determinations were made. The results are presented in table 5.

From the results presented in table 5, it appears that *Azotobacter*, in using the carbon of green manures, can fix a very small amount of nitrogen. In general the results agree with those of Dvorak (6). The very high amount of nitrogen assimilated in his experiment with green clover was never obtained in these experiments. This may be due in part to a difference in the strain of *Azotobacter* or to a difference in the composition of green tissue. However, it seems safe to assume that the carbohydrate in the green tissue available for this organism is small. Oat tissue apparently furnishes a better carbon source than clover.

Before proceeding to a summary of the results of these experiments, it is desirable to present a brief estimate of the possible value of green manures for nitrogen fixation under field conditions.

If an acre of soil 6 inches deep weighs 2,000,000 pounds, then the turning under of 30,000 pounds of green manure, or 1.5 per cent by weight of soil, should be followed by a nitrogen increase of 3 to 5 mgm. of nitrogen in 100 grams of soil, or 60 to 100 pounds of nitrogen in one acre of soil.

TABLE 5

Nitrogen assimilated by Azotobacter chroococcum in solution containing green clover and green oats

Incubated for 28 days at 26°C.

NO.	TREATMENT	NITROGEN IN 100 CC. OF SOLUTION					
		Clover			Oats		
		Total mgm.	Average mgm.	Increase mgm.	Total mgm.	Average mgm.	Increase mgm.
1	Sterile	23.5			16.8		
2	Sterile	23.7			16.8		
3	Sterile	23.7			16.8		
4	Sterile	23.7	23.7		16.8	16.8	
5	Sterile	23.7			16.7		
6	Sterile	23.5			16.9		
7	Sterile	23.9			16.7		
8	Sterile	23.9			16.7		
9	Azotobacter	24.6			17.8		
10	Azotobacter	24.4			17.9		
11	Azotobacter	24.3			17.6		
12	Azotobacter	24.3	24.4	0.7	17.6	17.7	0.9
13	Azotobacter	24.4			17.6		
14	Azotobacter	24.4			17.6		
15	Azotobacter	24.4			17.8		
16	Azotobacter	24.2			17.6		

SUMMARY

The results of all experiments show an increase in the assimilation of nitrogen due to the applications of green clover, wheat, or oats. This increase is noted both in solution and soil tests, and with pure cultures of *Azotobacter* growing in a medium containing green tissue.

Two kinds of soil were used in these experiments and in each case a small gain in nitrogen was observed. The variation in nitrogen content, in the untreated soil, is no doubt due to the error of analysis. The contents of separate plates were taken for analysis instead of portions of soil from the same container.

The green manures, probably because of the nature of their carbon content, stimulated nitrogen fixation very soon after treatment. It is interesting to note that in the majority of cases, the greatest gain resulted with the use of non-legume tissue as wheat and oats. These materials contained less nitrogen and more carbohydrate material, and decomposed more slowly than clover.

CONCLUSIONS⁸

1. From the data obtained under the conditions of the preceding experiments, it seems safe to conclude that green manures, as clover, wheat, or oats, when added to soil favor free nitrogen fixation.
2. A gain in nitrogen was noted in both soils. The increase was highest with field soil, due perhaps to a greater number of nitrogen-fixing organisms.
3. A gain in nitrogen was observed (a) where green tissue was applied to soil previously treated with a small amount of mannit; (b) where treated soil was used to inoculate Ashby's solution; and (c) when a pure culture of *Azotobacter* was used to inoculate a sterile solution containing green tissue.
4. The non-legume tissue stimulated fixation more than the legume, probably because of the nature of its carbohydrate content.
5. The results of these experiments in their entirety show that nitrogen fixation results from the addition of green manures to soil. Fortunately for the agriculturist, the plants giving best results are the ones lowest in nitrogen content.

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THE INFLUENCE OF FINENESS OF DIVISION OF PULVERIZED LIMESTONE ON CROP YIELD AS WELL AS THE CHEMICAL AND BACTERIOLOGICAL FACTORS IN SOIL FERTILITY¹

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REVIEW OF LITERATURE

If the value of an agricultural practice is determined by its age and universality, then that of liming must be well established, for the use of lime has been common to all lands since antiquity. Sound instinct has determined the use of lime and it is only the recent vogue of commercial fertilizers that has prevented liming from receiving the consideration which is its due. It is well known that the choice of one form of lime over another depends of necessity upon a variety of factors, such as soil texture, of organic matter content, items of cost, etc. It has been generally assumed in the past that burnt or slaked lime is more efficient than ground limestone for agricultural purposes. While much evidence has been offered to substantiate this claim, nevertheless, recent investigations have paved the way for the belief that limestone ground sufficiently fine may be considered as equivalent, if not superior, in value to the caustic forms of lime. Consequently, it is desirable to establish the possibilities and limitations of such contentions. To this end, one promising field of endeavor has furnished the basis for this investigation, namely, how do different grades of pulverized limestone influence crop yields, the chemical and bacteriological factors in soil fertility, and furthermore, how do they compare with the effects produced by burnt lime? More particularly the problem has been attacked from the following standpoints: the effect of different degrees of fineness of pulverized limestone on crop yields on different soils; the rate of neutralization of soil acidity; the bacteriological processes of ammonification, nitrification and nitrogen-fixation; the loss of limestone in drainage and the composition of the drainage water. While these may at first appear to be primarily scientific considerations, an attempt will be made to indicate their direct practical significance. The conclusions arrived at when taken in conjunction with other considerations may well furnish the basis for choice in the purchase of lime and therefore have an essential bearing upon the farmer's agricultural practice.

In approaching the problem concerning the influence of the different degrees of fineness of division of pulverized limestone, it is of interest to review the investigations pertinent to the subject at hand.

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Heinrich (39) appears to be the first investigator to report such data. The following results were recorded by him in 1908.

GRADES	PESAS	LUCERNE
GRAMS	GRAMS	GRAMS
1.5-2.0	32.0	17.1
1.0-1.5	38.8	22.7
0.5-1.0	43.3	28.9
0.5	48.0	28.9
No Ca + Fertilizer	22.6	0
No Ca + No fertilizer	26.0	0

He concludes from his work that the finer the grade of limestone the better the results obtained.

Voelcker (110) has compared "coarsely ground with finely ground" limestone and burnt lime when applied to the ordinary Woburn soil in the usual manner for the growth of wheat. Although the author does not state the degree of pulverization, nevertheless, the yields with the finely ground material are superior to those obtained with coarser, whether calcium or magnesium limestones were employed. Further, burnt lime was inferior to ground limestone whether coarse or fine. This result, Voelcker maintains, "is quite borne out by experience on the field scale in the Stackyard Field (plot 2bb) where applications of burnt lime have not shown good results at the outset in the case of wheat, but have required time to "work." This opens up the question as to whether the "alkalinity" of the burnt lime applied has had an influence in lowering the crop yield; also a further question as to whether the changes due to the use of lime may not be of a biological rather than a chemical nature." This interesting line of work is being prosecuted further by the author.

H. von Feilitzen (22) has compared slaked lime with pulverized limestone of the following grades: 0.2 mm., 0.2 to 0.5 mm., 0.5 to 1 mm., 1 to 2 mm. and 2 mm. He finds that slaked lime gives greater yields than calcium carbonate and both increase with an increase in fineness of division. The data, however, are not regarded as conclusive and the need for further experimentation is suggested.

Brehmer (8) treated a loam soil with limestone of different degrees of fineness (no mention being made of the degree of pulverization). The finer the material the greater was the increase in crop yield, while a mixture of 67 per cent CaCO_3 with 33 per cent CaO proved superior to the most highly pulverized limestone. In several instances in these experiments the increases between treatments were not greater than between duplicates receiving the same treatment.

Gerlach (35) supports Baessler's (4) contention that the solubility of calcium carbonate in marl increases proportionally with the amount of carbonate it

contains and the increased pulverization of the particles. The matter will bear further discussion in another connection.

The Rhode Island Agricultural Experiment Station (92) has published the early results of an experiment dealing with the growth of mangels and carrots when treated with limestone of varying degrees of pulverization. Their results are as follows:

TREATMENT	MANGELS, 1913	CARROTS, 1914
	pounds	pounds
No lime.....	107	175
10-20 Mesh limestone.....	138	314
20-24 Mesh limestone.....	204	469
40-80 Mesh limestone.....	389	563
Unsifted.....	279	470

The material coarser than 1/20 inch had little effect in the first year but considerable effect in the second. Whether this is due to a crop difference or one depending primarily upon increased availability of the limestone is considered questionable. It is evident, however, that in both cases an increase in fineness of division of pulverized limestone was paralleled by an increase in crop yield.

Thomas and Frear (104) have very recently published data indicating the importance of fineness of division in the use of pulverized limestone. The grades of limestone employed by them were as follows: 1/20 to 1/40 inch, 1/40 to 1/60 inch, 1/60 to 1/80 inch, 1/80 to 1/100 inch, and finer than 1/100 inch. Clover was grown in two soils held in paraffined baskets. The air-dry weights recorded indicate a progressive increase in yield as the limestone was more finely divided. This contention is limited by the fact that the authors obtained variations between soils receiving the same treatment as great as the differences between successive treatments. According to Frear the results of this investigation are being corroborated in a continuation series of experiments.

Ames and Schollenberger (3) determined the lasting effects of magnesian as compared with calcareous stone ground to different degrees of fineness. While the crop yields show as yet no marked differences, which is probably due to the high fertility of the soil employed, the authors concluded that the fineness of limestone is an important factor in determining its effectiveness. Material 100 per cent of which passes 10-mesh and 50 per cent of which passes 50-mesh and 35 to 40 per cent of which passes 100-mesh sieves is highly effective as a soil amendment; while material not finer than 1/20 inch produces but slight benefit in correcting soil acidity. Thus the results obtained in Ohio corroborate those of Pennsylvania, where it is recommended that ground limestone should pass at least a 60-mesh sieve.

Since the following experiments included a comparison of burnt lime with

the different grades of pulverized limestone in many instances, it is relevant at this point to consider briefly the previous work bearing upon this subject. Several of our agricultural experiment stations have given this matter considerable attention and especially noteworthy are the results obtained at Pennsylvania which cover a period of more than thirty years (30). The crushed limestone plats have given consistently higher yields than the plats which received the burnt lime dressing. An interesting point, established as a result of pot experimentation, is that on soils having a lime requirement (Veitch) over 1000 pounds of CaO per acre, ground limestone is manifestly superior to calcium oxide, while the differences on more alkaline soils are slight and more variable.

Patterson (85) also conducted field experiments for more than eleven years with caustic lime, by burning both stone and shells, and the carbonate of lime in ground shells and shell marl. The average crops of maize, wheat and hay were all larger on the plats treated with carbonate of lime.

Adams (1) states as a result of comparative work that no appreciable difference was noted in the benefit derived from an application of the same quantity of calcium oxide, whether in the form of ground limestone or in hydrated lime. In a recent bulletin of the Rhode Island Extension News Service a comparison of 80-mesh limestone with slaked lime shows that practically the same yield was obtained in each case.

Thorne (106) also has stated that "in actual practice, the experiments made by the Ohio Experiment Station have shown practically no superiority of one form of lime over another, provided the limestone has been so ground that 80 per cent of it will pass a sieve having 100 meshes to the linear inch, and provided also of course, that the two materials are used on the basis of actual calcium contained."

Saul (95) recommends finely ground limestone as superior to burnt lime, without furnishing experimental evidence to that effect.

Hardt (38) obtained larger yields with marl than with calcium oxide when applied in equivalent amounts to a heath soil under a five-year rotation with legumes every year.

Hoffman (41) gives the results of 79 experiments upon 150 farms in 17 regions of the German Empire. In most cases burnt lime was beneficial. On light soils poor in humus and in dry seasons the carbonate was superior to the quick lime.

Meyer (73) grew mustard and beans alternately for four years in pots of sandy loam treated with different forms of lime. As a result of this experiment, the yields resulting from the application of ground limestone and calcium carbonate (c.p.) were calculated as 100 and the burnt lime received then a valuation of only 95. The ground limestone had the following mechanical analysis: 74.3 per cent of flour (equivalent to no. 100 Thomas slag sieve), 21.4 per cent finer than 0.5 mm., and 4.3 per cent larger than 0.5 mm. but smaller than 10 mm.

Mooers (76) reports comparative experiments with ground limestone and caustic lime conducted for four years with a variety of crops including corn, wheat, oats, barley, sorghum, clover, grass, cowpeas and cotton. He concludes that "in these trials two tons of ground limestone proved somewhat superior to one ton of burnt lime."

Voorhees, Lipman and Brown (113) concluded, with certain reservations, from their investigations of the comparative value of different forms of lime that "the carbonate led to the production of more dry matter, more nitrogen and more ash than did the burnt lime."

Porter and Gaut (91) conclude from their comparison of cob (kiln) lime, ground lime and ground limestone as top-dressings for various pastures, with and without manure, that ground limestone was more profitable for grass lands in need of lime than CaO.

Yokayama (124) maintains as a result of experiments with oats grown in sand cultures that precipitated calcium carbonate is far more effective than fine limestone meal in increasing crop yield.

Ellett (19) emphasizes the importance of relative cost in calculations concerning the use of different forms of lime and maintains that ground limestone gives results as good as those obtained with burnt lime.

H. von Feilitzen (21) found that slaked lime gave greater yields than carbonate, but the data are not conclusive. In a later publication (22) the same author reported results obtained showing that calcium carbonate was as valuable as burnt lime on acid soils poor in lime.

Wright (123) in investigations conducted on medium and light soils found that in the beginning burnt lime was superior to gas lime, but that in the seven succeeding years of the rotation, the latter was much more beneficial. In that period it gave a total increase of more than three times the value of that produced by burnt lime and because of the lower cost proved much more profitable.

Riggl (93) found that calcium carbonate as a top-dressing was preferable to burnt lime except where the latter was used in very small amounts. He is of the opinion that burnt lime is better than calcium carbonate because of the physical and bacteriological effects resulting from the finer state of division of the particles.

Voelcker (111) states as a result of some grass experiments that "it is still early for the application to tell, but at the present time if any plots were selected for choice they would be plots 4 and 5 where ground limestone and ground chalk, respectively, have been used. Ground lime also seems to have done better than lump lime."

Barker and Collison (5) discuss the superiority of ground limestone over burnt lime, and, without publishing data, conclude that 10-mesh limestone is to be recommended.

Rothert (94) in noting the depressing effect upon crop yield of increased quantities of burnt lime find that on a loam soil barley gave a greater yield

with 0.5 per cent of calcium carbonate than with 0.25 per cent of burnt lime. In sand the reverse was true.

Lauder, Fagan and Steele (58) record the beneficial effects of ground lime and waste carbonate for finger and toe disease. They state that when the latter product is available it may be employed instead of burnt lime with satisfactory results.

Grantham (36) in a preliminary report asserts that "results thus far indicate that the carbonate of lime is practically as effective as the burned forms of lime in promoting the decomposition of organic matter and the availability of nitrogen."

The effect upon soil reaction of applications of limestone has been the subject of investigation by the Pennsylvania Agricultural Experiment Station (29, 117, 118). In this connection it may be stated that Dr. W. Frear has contributed one of the most valuable and comprehensive surveys of the literature dealing with liming (30). Because of the thoroughness of his recent bulletin it has been unnecessary in the present instance to do more than briefly indicate the nature of the problems attacked by various investigators concerning pulverized limestone. A more complete discussion will be found in the above-mentioned bulletin which has furnished material for this bibliography. In an experiment on nitrification, White (118) found that at the end of four weeks an increase in fineness of division of limestone used was responsible for a corresponding decrease in the lime-requirement of the soil. The influence of crops upon soil reaction has also been an interesting point established by Gardner and Brown (34) in plot experiments with white clover. In general it was found that the soils of the unlimed plots showed an increase in acidity on the average after the crop roots had been removed, and even those limed or treated with fine carbonate of lime in quantity sufficient to neutralize the original acidity were found to be acid to a slight degree after the crop was removed.

Veitch (109) has observed that different plants affect the soil reaction differently.

Lipman and Blair (64) found in the course of field experiments that after a number of years plots (including one series unlimed, the other treated with limestone) were acid; those of the unlimed groups requiring from 1000 to 2000 pounds (average 1840 pounds) of lime per acre, while those of the limestone-treated group averaged about 700 pounds. Volunteer clover sprang up on most of these plots, but its yield on the limed plots was double that on the unlimed.

The beneficial effect of ground limestone upon the growth of legumes has been observed by the following investigators: Bäessler (4), Hardt (38), Lipman (63), Stevenson and Weston (100), Peter and Jones (88) and many others.

Various technical phases as well as general discussions pertaining to ground limestone have been treated in an interesting manner by Ames (2), Bernard

(6), P. E. Brown (14), Dumont (18), Fippin (23), Fischer (25), Frear (30), Frear and Thomas (31), Johnston (50), Morse (77), Neuffer (81), Pfeiffer (90), Simmermacher (99), Strebust (101), Takeuchi (103), Wheeler (115) and Whitson and Weir (119).

THE EFFECT OF FINENESS OF DIVISION OF PULVERIZED LIMESTONE ON CROP YIELD, NITROGEN CONTENT AND LIME-REQUIREMENT OF VARIOUS SOILS

It is apparent from the foregoing review that there is a paucity of experimental evidence concerning the different grades of pulverized limestone and their relative value compared with the burnt forms of lime. The practical significance which attaches to this problem is responsible for the following greenhouse experiments which were begun in the summer of 1914. Crimson clover was grown on soils which may be regarded as typical of rather extensive fertile areas in the United States. These were respectively designated as Carrington (Iowa) silt loam, Wooster (Ohio) silt loam, Cumberland (Tennessee) silt loam, Norfolk (Virginia) sandy loam, Sierra (California) sandy loam and Portsmouth (New Jersey) acid muck. They were obtained through the courtesy of Dr. P. E. Brown, Prof. C. E. Thorne, Prof. C. A. Mooers, Dr. J. W. Schoene and Dr. J. G. Lipman. The procedure was to fill 10-pound glazed earthenware pots with soil to which was added 2 grams of acid phosphate (14 per cent P_2O_5) and 1 gram of potassium chloride (50 per cent K_2O). (All soils used in this and following experiments were from unlimed and unfertilized plots and were air-dried, thoroughly mixed, and sieved through a 12-mesh sieve.) Limestone was then added which had been pulverized and hand-sieved as follows: 20 to 40-mesh, 60 to 80-mesh, 100 to 200-mesh and finer than 200-mesh, as well as calcium oxide (c.p.) which had been ground in a mortar. The limestone used was a uniform crystalline grey limestone having the following composition:

	per cent
SiO_2	3.75
R_2O	1.72
$CaCO_3$	92.00
$MgCO_3$	3.45

The lime was added in quantities sufficient to neutralize the soil acidity as determined by the Veitch method. The lime-requirements of the different soils in pounds of CaO per acre were as follows: Carrington silt loam, 3700; Wooster silt loam, 4000; Cumberland silt loam, 3400; Norfolk sandy loam, 2300; Portsmouth acid muck, 8733; and Sierra sandy loam, alkaline. Thirty tested seeds of crimson clover were planted in each pot on February 10. The plants were thinned to 15 in number by April 15 and the pots kept at optimum temperature and moisture conditions, being made up to weight twice weekly. The crop was harvested June 29. During the warm weather thrips attacked the plants and frequent spraying with nicotine sulfate became neces-

sary. However, the crops were not unduly injured. The results of the crop yield (dry matter), per cent of nitrogen, nitrogen content in grams, and original and final lime-requirements are recorded in tables 1 and 2.

TABLE 1

The effect of fineness of division of pulverized limestone on crimson clover and lime-requirement

TREATMENT	WEIGHT OF CROP	DUP. & AVERAGE	NITROGEN	AVERAGE NITROGEN	TOTAL NITROGEN	ORIGINAL LIME-REQUIREMENT OF CaO PER A	LIME - REQUIREMENT AT END OF CaO PER A	DUP. & AVERAGE OF CaO PER A	AVERAGE LIME-REQUIREMENT OF CaO PER A
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Norfolk sandy loam

	gms.	gms.	gms.	per cent	per cent	per cent	gm.	pounds	pounds	pounds
Check (Minerals).....	17.9	18.9	18.4	1.19	1.24	1.22	0.22	2,300	2,500	2,600
5.9 grams, 20-40 mesh...	18.9	22.2	20.6	1.63	1.75	1.69	0.35	2,300	1,600	1,700
5.9 grams, 60-80 mesh...	23.7	26.9	25.3	2.04	2.06	2.05	0.52	2,300	1,000	1,300
5.9 grams, 100-200 mesh.	23.3	26.0	24.7	2.04	2.06	2.05	0.51	2,300	1,000	1,000
5.9 grams, 200 + mesh...	26.5	27.3	26.9	2.10	2.09	2.10	0.56	2,300	700	1,000
3.3 grams, CaO.....	29.1	30.0	29.6	2.00	2.00	2.00	0.59	2,300	900	1,000

Wooster silt loam

	gms.	gms.	gms.	per cent	per cent	per cent	gm.	pounds	pounds	pounds
Check (Minerals).....	4.9	5.6	5.3	1.48	1.37	1.43	0.08	4,000	4,100	4,000
10.9 grams, 20-40 mesh..	7.3	10.4	8.9	1.86	1.88	1.87	0.35	4,000	2,700	2,700
10.9 grams, 60-80 mesh..	18.1	20.5	19.3	1.97	1.93	1.95	0.38	4,000	100	300
10.9 grams, 100-200 mesh	21.0	23.4	22.1	1.86	1.75	1.81	0.40	4,000	200	200
10.9 grams, 200 + mesh..	20.9	21.8	21.4	1.72	1.81	1.77	0.38	4,000	400	600
6.1 grams, CaO.....	20.3	21.9	21.1	1.64	1.67	1.66	0.35	4,000	200	200

Carrington silt loam

	gms.	gms.	gms.	per cent	per cent	per cent	gm.	pounds	pounds	pounds
Check (Minerals).....	26.9	18.8	22.9	2.01	2.00	2.01	0.46	3,700	3,800	3,700
10.0 grams, 20-40 mesh..	28.0	30.2	29.1	1.89	1.85	1.87	0.54	3,700	2,400	2,800
10.0 grams, 60-80 mesh..	26.3	30.1	28.2	2.06	2.12	2.09	0.59	3,700	1,200	1,900
10.0 grams, 100-200 mesh	25.6	32.9	29.3	2.17	2.14	2.16	0.64	3,700	100	100
10.0 grams, 200 + mesh..	31.5	33.6	32.6	2.17	2.21	2.19	0.71	3,700	100	100
5.6 grams, CaO.....	30.7	39.5	35.1	2.04	2.03	2.04	0.72	3,700	300	500

Cumberland silt loam

	gms.	gms.	gms.	per cent	per cent	per cent	gm.	pounds	pounds	pounds
Check (minerals).....	7.6	7.7	7.7	1.41	1.41	1.41	0.11	3,400	4,000	4,000
9 grams, 20-40 mesh....	10.0	10.6	10.3	1.48	1.42	1.45	0.15	3,400	2,700	2,700
9 grams, 60-80 mesh....	24.7	12.7	18.7	2.07	2.04	2.06	0.39	3,400	1,900	1,500
9 grams, 100-200 mesh....	18.8	19.4	19.1	1.92	1.93	1.93	0.37	3,400	1,100	1,100
9 grams, 200 + mesh....	19.7	20.7	20.2	2.00	2.04	2.02	0.41	3,400	900	700
5 grams, CaO.....	13.3	18.6	16.0	2.05	2.04	2.05	0.33	3,400	1,100	1,600

TABLE 2

The effect of fineness of division of pulverized limestone on crimson clover and lime-requirement

TREATMENT	WEIGHT OF CROP	DUPLI- CATE	AVER- AGE	NITRO- GEN	NITRO- GEN	AVER- AGE NITRO- GEN	TOTAL NITRO- GEN
<i>Sierra sandy loam</i>							
Check (Minerals).....	32.3	37.0	34.7	2.06	2.01	2.04	0.71
2.6 grams, 20-40 mesh.....	37.0	41.5	39.8	1.94	1.92	1.93	0.77
2.6 grams, 60-80 mesh.....	44.6	45.3	45.0	1.92	1.90	1.91	0.86
2.6 grams, 100-200 mesh.....	36.5	38.0	37.2	1.68	1.70	1.69	0.63
2.6 grams, 200 + mesh.....	32.7	33.0	32.9	1.93	1.88	1.91	0.63
1.46 grams, CaO.....	31.0	27.8	29.4	1.80	1.77	1.79	0.53
<i>Portsmouth acid muck</i>							
Check (minerals).....	0.2	0.3	0.3	2.42	2.43	2.43	0.01
2 grams, 20-40 mesh.....	18.9	18.5	18.7	1.66	1.62	1.64	0.31
2 grams, 60-80 mesh.....	21.9	22.0	22.0	2.04	2.10	2.07	0.46
2 grams, 100-200 mesh.....	23.7	23.9	23.8	2.12	2.14	2.13	0.51
2 grams, 200 + mesh.....	22.9	25.1	24.0	2.24	2.20	2.22	0.53
Commercial, 200 mesh.....	23.2	24.2	23.7	2.10	2.04	2.07	0.49

Discussion

To facilitate the discussion of the results recorded in tables 1 and 2, a summary is presented in table 3.

Table 3 shows the relative effect of fineness of division of pulverized limestone and burnt lime on crop yield on five typical soils. It will be observed that in the case of the Norfolk sandy loam an increase in fineness of division was accompanied (with one exception) by an increase in crop yield. The 200-mesh gave a yield almost one-third greater than the 20-mesh limestone. On this soil burnt lime proved superior to even the finest grade of limestone. The Wooster silt loam likewise exhibits an increase in crop yield with an increase in fineness of division of pulverized limestone up to the finest grade. It will be noted that the burnt lime, as well as the latter, falls below the yield obtained with 100-mesh limestone. This may be interpreted as indicating that the 200-mesh limestone and burnt lime were especially active in the neutralizing of the soil acidity at the outset of the experiment, but permitted a greater subsequent accumulation of acidity. The Carrington silt loam responded to an increase in fineness of division of pulverized limestone which was responsible for an increase in crop yield (with one exception). The burnt lime proved superior to all grades of limestone on this heavy soil rich in organic matter. The Cumberland silt loam otherwise shows very strikingly that an increase in fineness of division of pulverized limestone is accompanied by an increase in crop yield. In this case the 200-mesh proved to be doubly

as efficient as the 20-mesh limestone. The burnt lime was slightly inferior to the former. Thus, averaging the results for these soils in the last column of table 3, it will be seen that with an increase in fineness of division of pulverized limestone there was a proportional increase in crop yield. Furthermore the 200-mesh proved to be fully one-third again as valuable as the 20-mesh limestone. Practically all the forms of lime gave double the yield

TABLE 3

Summary showing relative effect of fineness of division of pulverized limestone and CaO on the crop yield, total nitrogen in grams, and decrease in per cent of lime-requirement of five typical soils

TREATMENT	NORFOLK SANDY LOAM	WOOSTER SILT LOAM	CARRINGTON SILT LOAM	CUMBER- LAND SILT LOAM	SERRA SANDY LOAM*	AVERAGE
<i>Crop yield</i>						
Check.....	100	100	100	100	100	100
20-40 mesh.....	112	357	127	134	115	183
60-80 mesh.....	138	364	123	243	130	217
100-200 mesh.....	134	419	130	248	108	233
200 + mesh.....	146	404	142	262	95	239
CaO.....	161	398	153	208	85	230
<i>Total nitrogen in grams</i>						
Check.....	100	100	100	100	100	100
20-40 mesh.....	159	438	117	136	108	213
60-80 mesh.....	236	475	128	355	121	299
100-200 mesh.....	231	500	139	336	89	302
200 + mesh.....	255	475	154	373	89	314
CaO.....	268	438	157	300	75	291
<i>Decrease in lime-requirement</i>						
Check.....	100	100	100	100		100
20-40 mesh.....	135	134	132	133		134
60-80 mesh.....	154	195	158	158		166
100-200 mesh.....	162	195	197	173		181
200 + mesh.....	165	188	197	180		183
CaO.....	162	151	189	165		167

* Not averaged.

obtained on the unlimed soils. There is little choice between the yields obtained with burnt lime and 200-mesh limestone since each was superior in two of the four instances. Obviously, the chemical and physical nature of the soil, together with its content of organic matter, are the determining factors in the interpretation of such results.

The Sierra sandy loam which was originally alkaline presented a peculiar phenomenon, namely, the coarser limestones which act more slowly gave an

increased crop yield while the finer material and burnt lime were responsible for a decrease. The interpretation which many placed upon these facts is that the more rapidly-acting forms of lime cause certain chemical transformations to take place in the soil which prove unfavorable to crop growth. The results obtained with Portsmouth acid muck are presented in table 2. Here again it is evident that an increase in fineness of division of pulverized limestone is accompanied by an increase in crop yield. It is interesting to note that with the commercial product used ("Edison Pulverized Limestone," 80 per cent of which is guaranteed to pass a 200-mesh sieve) the crop yield was identical with that obtained with 100 to 200-mesh material, thereby fulfilling its guarantee by crop yield as well as mechanical analysis.

The importance of the nitrogen content of a leguminous crop is universally recognized alike for its feeding value and its fertilizing value in a cover crop. Summary table 3 shows the relative effect of fineness of division of pulverized limestone and burnt lime on the total nitrogen (in grams) of crimson clover. It is apparent that in practically all instances the crop yield is paralleled by the total nitrogen content; that is, with an increase in fineness of division of pulverized limestone there is an increase in the total nitrogen content of the crop. This is well illustrated in figure 1.

Undoubtedly, a highly essential consideration in the choice of the different forms of lime (aside from the paramount one of cost) is the power to neutralize soil acidity. In the present experiment, the Veitch lime-requirement of the various soils was determined at the outset, as previously recorded. After the crops had been harvested representative samples were obtained from each pot and the lime-requirement again determined. In this manner it was possible to obtain an index as to the reduction in the lime-requirements of these soils by the different forms of lime employed. While the Veitch method is open to objection for this purpose because of the fact that any residue of limestone would be soluble to the degree of its fineness and thus artificially weight the results, nevertheless, this method was adopted since it is so widely used and great accuracy is not imperative. It is, then, interesting to note that with but few exceptions an increase in fineness of division of pulverized limestone is responsible for a proportional decrease in the lime-requirements of the soils under consideration. As shown in table 3, the 200-mesh proved to be two and one-half times as effective as either the 60-mesh limestone or burnt limestone. The explanation previously suggested may be offered again in this connection, namely, the burnt lime may have been much more efficient in reducing the lime-requirement initially, but that it was used up so rapidly as to allow a subsequent development of acidity. In general the decrease in lime-requirement as a result of fineness of division is quite closely correlated with crop yield and total nitrogen content as shown in figure 1.

Thus the points which have been established in this experiment may be summarized as follows:

1. Soils representative of extensive fertile areas namely, Norfolk and Sierra sandy loams, Carrington, Wooster and Cumberland silt loams and Portsmouth acid muck, when growing crimson clover in pots, produced similar results when treated with limestone pulverized 20 to 40, 60 to 80, 100 to 200, finer than 200-mesh and burnt lime (c.p.).

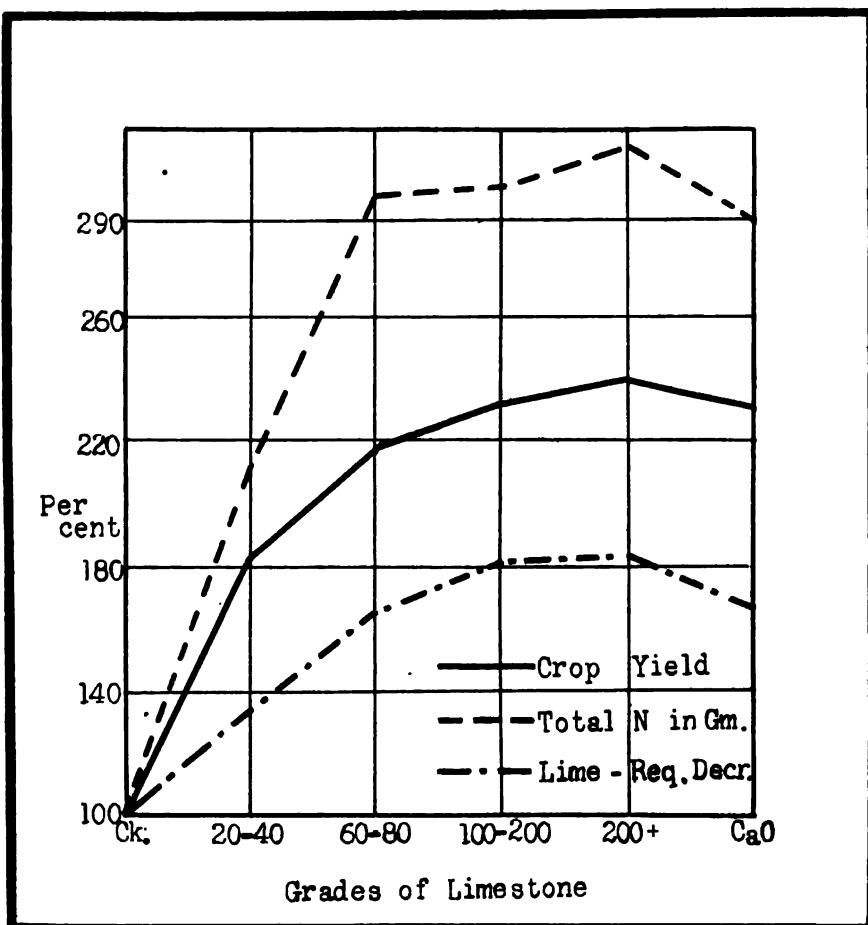


FIG. 1. DIAGRAM SHOWING THE EFFECT OF DIFFERENT DEGREES OF NITROGEN, IN GRAMS, AND THE DECREASE IN LIME-REQUIREMENT OF SEVERAL SOILS, EXPRESSED IN PER CENT

2. An increase in fineness of division of pulverized limestone was accompanied by a proportional increase in crop yield and total nitrogen content and a corresponding decrease in lime-requirement, a close correlation existing between these factors.

3. Two-hundred-mesh proved one-third again as effective in influencing crop yield as 20-mesh limestone, and more than twice as effective in reducing the lime-requirement of soils.

4. There was little choice between burnt lime and 200-mesh limestone, though in general the latter appeared preferable. Burnt lime proved superior to coarse limestone.

THE RATE OF NEUTRALIZATION OF SOIL ACIDITY AS INFLUENCED BY DIFFERENT DEGREES OF FINENESS OF PULVERIZED LIMESTONE

In view of the enhanced effects accruing from the increased fineness of division of pulverized limestone, the practical question arose regarding the value of the different grades of pulverized limestone and burnt lime with regard to the rate of neutralization of soil acidity. Obviously, this is of great significance to the farmer who is seeking to neutralize the soil acidity so that a crop may be planted without undue delay. Since no previous work bearing directly upon this point could be found it became necessary to devise a method for measuring the rate of neutralization of acidity in soil treated with different grades of limestone and burnt lime. The Veitch method hardly lent itself to this purpose because of the fact that the water-bath treatment would result in different quantities of residual limestone going into solution, thereby invalidating any comparative results. Therefore, it was deemed advisable to measure the rate of neutralization by the carbon dioxide evolved. This was accomplished by adding a known amount of limestone to the soil (sufficient to satisfy the Veitch lime-requirement) which was calculated to its carbon dioxide content. At certain intervals the carbon dioxide remaining in the soil was determined, the loss of carbon dioxide compared with the original carbon dioxide content representing the amount of limestone which had been used up in neutralizing the soil acidity. The method employed is virtually a modification of that used by Tacke (92) and Truog (108), the apparatus being arranged as in figure 2.*

The procedure was as follows:

Ten grams of soil were placed in the 250-cc. Florence Flask (*A*) and 15 cc. of HCl (1: 3) run in through the separatory funnel (*B*). Enough boiling water was then added to make up a volume of about 75 cc. and the solution heated up to the boiling point in two minutes and kept there for one minute. The heat applied with a Bunsen flame was regulated in such a manner as to keep the rate at which the bubbles passed through the tower of beads (*D*) approximately constant and not very rapid. The tower (*D*) held a measured quantity of Ba(OH)₂ and a few drops of phenolphthalein, as did the 200-cc. Erlenmeyer flask at *E* which served as a safety flask for any carbon dioxide which might not be held at *D*. After heating for three minutes the pinchcock at *C* was closed and the flask *A* filled with boiling water from an inverted wash bottle. In order to do this properly it was usually necessary to suck on the rubber tube at *G* while cautiously opening the pinchcock at *C*. Any sudden changes in room temperature or air currents from open windows, etc., must be guarded against. When the water mounted to *F* the stop-cock at that point was closed; thus all the carbon dioxide present in the Florence flask has been displaced by boiling water. The barium hydrate solution in *D* (standardized so that 1 cc. was equivalent to 0.23 mgm.

* The writer is indebted to Mr. R. E. Curtis for his valuable suggestions in this connection.

of CO_2) was then emptied into a funnel placed in a 500-cc. Erlenmeyer flask. The beads remained on the funnel and were thoroughly washed with boiling water, as was the chamber *D* and the flask *E*, the contents of which also had been emptied. The solution of $\text{Ba}(\text{OH})_2$ was then titrated with oxalic acid equal in strength to the $\text{Ba}(\text{OH})_2$. Thus the amount of $\text{Ba}(\text{OH})_2$, which was used in absorbing the carbon dioxide evolved was determined and the latter calculated accordingly. It might be mentioned that when small quantities of $\text{Ba}(\text{OH})_2$ were placed in *D* enough CO_2 -free water was added to bring the volume just below the connecting tube, and a small quantity of water was added also to *E*, so that the tube dipped below the liquid. The ease in manipulation and the fact that it required only ten to fifteen minutes to complete a determination were primary considerations in the adoption of this method. The duplicates checked quite closely.

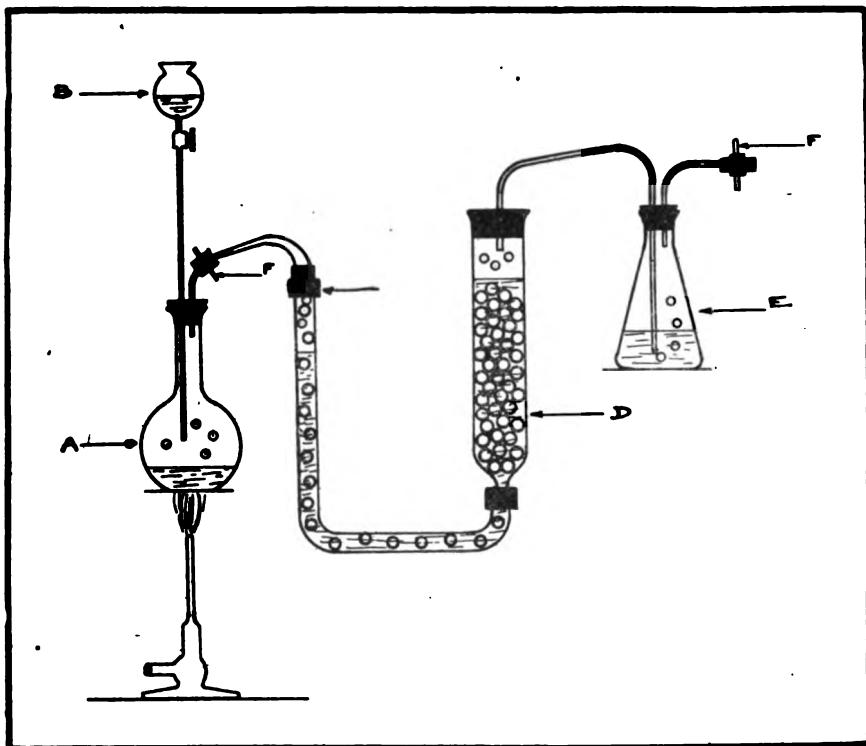


FIG. 2. DIAGRAM OF APPARATUS USED IN DETERMINING CARBON DIOXIDE IN SOIL

The following investigators have considered the solubility of calcium carbonate. Cameron and Bell (15), Holleman (43), Le Blanc and Novotny (59), Schloessing (97), Treadwell and Reuter (107), Warington (114), Whipple and Mayer (116), while Gerlach (35) has compared the solubility of limestone pulverized to different degrees of fineness. With regard to the difference in the number of particles per cubic foot of limestone when pulverized to different grades, the following data have been obtained through the courtesy of Dr. H. E. Kieffer. One cubic foot of limestone will make 64,000,000 pieces which will

passes a 12-mesh sieve, and 512,000,000,000 pieces which will pass a 200-mesh sieve; thus the finer grinding in this case means 8000 times the number of particles obtained with the coarser grinding. Or calculating it in another way, 1 ton of 12-mesh limestone when applied to the soil would give 17,337 pieces for each square foot, while 1 ton of 200-mesh limestone would give 13,869,051 pieces per square foot. The importance of this both from the point of uniform distribution and increased solubility cannot be over-estimated.

The influence of calcium carbonate on organic matter has been the subject of investigations by Frear (30), Kossowitch (55), Lemmerman (60), Peterson (89) and Wollny (121). It is evident that the organic matter of the soil would be attacked in varying degrees by different grades of pulverized limestone.

The present experiment was carried out first with Norfolk sandy loam and Penn clay loam and later with Sassafras sandy loam. Five-pound glazed earthenware pots were filled with 1500 grams of Norfolk sandy loam and 1200 grams of Penn clay loam, respectively. The lime-requirement of the former, according to the Veitch method, was 2200 pounds and the latter 1100 pounds of CaO per acre. Limestone of the different grades previously referred to and burnt lime (c.p.) were added in sufficient amount to neutralize the lime-requirements of the soils to be treated. The amount of limestone added was calculated on the basis of 90 per cent purity. Each treatment was carried out in duplicate, and untreated soils served as checks. The pots were tared, filled with the proper quantity of soil, 20 cc. of a soil infusion (known to contain the more important soil organisms in abundance) were added and enough distilled water to bring the soils up to their respective optimum moisture contents, namely, 13 per cent for the Norfolk sandy loam and 26 per cent for the Penn clay loam. The pots were supplied with glass covers to prevent evaporation and incubated in a dark room at 20°C. for 7 weeks. Moisture was added as required to the soils uniform throughout the duration of the experiment. At the end of 3 weeks, the soil in each pot was carefully mixed and a representative sample of 100 grams removed, dried and analysed for carbon dioxide content by the method previously described. The same procedure was repeated at the end of 7 weeks.

In table 4 will be found the amounts of CO₂ present, in the soils variously treated at the end of 3 weeks. The last column of the table was calculated in the following manner. The amount of limestone applied per 10 grams of Norfolk sandy loam was equivalent to 5.7 mgm. of CO₂. The check soils indicated the presence of 3.8 mgm. of CO₂, therefore, the total CO₂ content of each soil was 9.5 mgm. The CO₂ found to be present in 10 grams of soil in each pot after 3 weeks was then subtracted from 9.5 mgm. of CO₂ and the remainder recorded the mgm. of CO₂ used (calcium carbonate) in neutralizing soil acidity. It is evident from the results secured with both Norfolk sandy loam and Penn clay loam that an increase in fineness of division of pulverized limestone was accompanied by a proportional increase in the amount of calcium carbonate used up in neutralizing the acidity of the soil. The chief

point of interest, however, lies in the fact that the limestone finer than 60-mesh had neutralized the soil acidity at the end of 3 weeks, while this was not the case with the 20-mesh product. It appears that the finer grades of limestone, as well as the burnt lime (which had also neutralized the soil acidity in this period of time) had begun to attack the organic matter of the soil.

The results obtained at the end of 7 weeks as shown in table 5 indicated that the coarse limestone had very nearly succeeded neutralizing the soil acidity in the Norfolk sandy loam but was not quite so effective in the Penn clay loam. The other grades of limestone exhibited practically no differences other than those already noted. Thus from the foregoing data it appears that the finer grades of limestone neutralize the soil acidity in 3

TABLE 4

The effect of fineness of division of pulverized limestone on the neutralization of acidity as measured by CO₂ present after three weeks' incubation

TREATMENT	CO ₂	CO ₂	AVERAGE CO ₂	INCREASE OVER CHECK CO ₂ USED
<i>Norfolk sandy loam</i>				
Check.....	3.7	3.9	3.8	
20- 40 mesh.....	6.3	7.5	6.9	2.6
60- 80 mesh.....	3.1	3.2	3.2	6.3
100-200 mesh.....	3.0	2.3	2.7	6.8
200 + mesh.....	1.6	1.8	1.7	7.8
CaO.....	1.4	1.6	1.5	8.0
<i>Penn clay loam</i>				
Check.....	4.6	4.7	4.7	
20- 40 mesh.....	6.9	7.3	7.1	1.4
60- 80 mesh.....	5.6	5.5	5.6	2.9
100-200 mesh.....	4.1	4.0	4.1	4.4
200 + mesh.....	3.2	3.9	3.6	4.9
CaO.....	2.8	2.4	2.6	5.9

weeks even in soils of such widely divergent types as those employed in the present instance. In this respect they proved to be as efficient as burnt lime. The coarser limestone, on the other hand, required more than double this period of time to neutralize the soil acidity. Wilkins (120), in unpublished data, has shown that in a given period of time an increase in fineness of division of pulverized limestone is responsible for an increased production of CO₂ and nitrates.

In order to obtain further corroboration of the points brought out in the preceding experiment, the latter was repeated under similar conditions with another soil, namely, Sassafras sandy loam having a lime-requirement of 3300 pounds of CaO per acre. An additional feature was the use of two and one-

half times the amount of 20-mesh limestone as required in order to determine whether larger amounts of coarser limestone would be as effective as smaller amounts of finer material in a short period of time.³ This seemed advisable in view of the fact that the pot experiments previously referred to indicated that the 200-mesh limestone was most effective in neutralizing acidity and further because the same proportion was maintained with regard to the rate of time required for neutralization.

TABLE 5

The effect of fineness of division of pulverized limestone on the neutralization of acidity as measured by CO₂ present after seven weeks' incubation

TREATMENT	CO ₂	CO ₂	AVERAGE CO ₂	INCREASE OVER CHECK CO ₂ USED
<i>Norfolk sandy loam</i>				
Check.....	2.4	2.7	2.6	
20- 40 mesh.....	3.5	4.4	4.0	4.3
60- 80 mesh.....	3.0	2.6	2.8	5.5
100-200 mesh.....	1.7	1.8	1.8	6.5
200 + mesh.....	1.8	2.1	2.0	6.3
CaO.....	1.6	1.6	1.6	6.7
<i>Penn clay loam</i>				
Check.....	3.8	3.7	3.8	
20- 40 mesh.....	6.0	5.8	5.9	1.7
60- 80 mesh.....	4.2	4.7	4.5	3.1
100-200 mesh.....	2.6	2.9	2.8	4.8
200 + mesh.....	2.6	2.9	2.8	4.8
CaO.....	1.7	1.7	1.7	5.9

The results as recorded in table 6 are substantially the same as those obtained with the other soil types, namely, with an increase in fineness of pulverized limestone there is an increased amount of calcium carbonate used in neutralizing soil acidity. Concerning the salient point of this investigation it is apparent that the 200-mesh has required but two weeks to effect neutralization, the 60-mesh requires three to four weeks, while the 20-mesh limestone requires a still longer period of time. The burnt lime, as might be anticipated, neutralized the soil acidity in the first week and must be considered as more rapid in its activity than even the finest grade of limestone used. Glancing at the results obtained with the increased quantity of coarse limestone, it will be noted that it proved fully as effective as the finest limestone.

Thus the preference for finer over coarser limestone would resolve itself chiefly to a question of economy in purchase, hauling, distribution, etc. The

³ The writer is indebted to Mr. C. M. Haensler for suggesting this point.

TABLE 6

The rate of neutralization of acidity by different degrees of fineness of pulverized limestone in Sassafras sandy loam

TREATMENT	CO ₂	CO ₂	AVERAGE CO ₂	CO ₂ USED
<i>After one week</i>				
Check.....	1.6	2.0	1.8	
20-40 mesh.....	5.9	6.4	6.2	5.1
2.5 × 20-40 mesh.....	16.2	16.7	16.5	7.2
60-80 mesh.....	5.9	6.4	6.2	5.1
100-200 mesh.....	5.7	5.9	5.8	5.5
200 + mesh.....	5.0	4.8	4.9	6.4
CaO.....	4.3	4.1	4.2	7.1
<i>Second week</i>				
Check.....	1.6	2.1	1.9	
20-40 mesh.....	6.8	7.0	6.9	4.5
2.5 × 20-40 mesh.....	15.2	15.3	15.3	8.4
60-80 mesh.....	5.5	5.9	5.7	5.7
100-200 mesh.....	4.6	4.5	4.6	6.8
200 + mesh.....	2.6	2.9	2.8	8.6
CaO.....	2.6	2.8	2.7	8.7
<i>Third week</i>				
Check.....	1.9	1.9	1.9	
20-40 mesh.....	7.0	7.4	7.2	4.2
2.5 × 20-40 mesh.....	15.7	15.0	15.4	8.5
60-80 mesh.....	5.5	5.2	5.4	6.0
100-200 mesh.....	4.6	5.2	4.9	6.5
200 + mesh.....	4.3	3.8	4.1	7.3
CaO.....	2.9	3.1	3.0	8.4
<i>Fourth week</i>				
Check.....	1.0	1.0	1.1	
20-40 mesh.....	6.5	6.4	6.5	4.1
2.5 × 20-40 mesh.....	15.0	14.7	14.9	8.8
60-80 mesh.....	5.2	5.1	5.2	5.4
100-200 mesh.....	3.4	3.5	3.5	7.1
200 + mesh.....	2.5	2.7	2.6	8.0
CaO.....	2.6	2.6	2.6	8.0

points established in the preceding experiments may be summarized as follows:

1. An apparatus is described for measuring the rate of neutralization of acidity by different degrees of fineness of pulverized limestone.

2. It was found that with Norfolk sandy loam as well as with Penn clay loam an increase in fineness of division of pulverized limestone was responsible for an increase in amount of limestone used up in the neutralization of soil acidity.

3. At the end of three weeks the limestone finer than 60 to 80-mesh had neutralized the acidity in both soils, while 20 to 40-mesh limestone required something more than seven weeks to accomplish the same result. The finer mesh limestone was as effective in three weeks as burnt lime.

4. With Sassafras sandy loam, the above results were corroborated by weekly determinations. In this soil the burnt lime neutralized the acidity in one week, the 200-mesh limestone required two weeks, the 60-mesh three to four weeks and the 20-mesh even a longer period of time.

5. Twenty-mesh limestone used in quantities two and one-half times as great as fine limestone proved fully as effective as the latter in three weeks.

THE FINENESS OF DIVISION OF PULVERIZED LIMESTONE AS INFLUENCING THE BACTERIOLOGICAL ACTIVITIES IN THE SOIL

No study of a soil amendment could be considered adequate without an investigation concerning its effect upon the activities of the soil microorganisms. This is especially true of lime since it determines to such a marked degree the reaction of the soil. So far as the writer has been able to ascertain only one experiment has been reported which deals with the effect of fineness of division of pulverized limestone upon soil biological activities. This work has been done at the Pennsylvania Agricultural Experiment Station (117), where a study was being made of the effects of pure and magnesian limestone upon the formation of nitrate as compared with equivalent amounts of burned lime.⁴ It was found that an increase in the fineness of division of pulverized limestone caused an increase in nitrification. Burnt lime proved superior to any grade of limestone used.

The fact that in general lime increases the number of soil organisms has been observed by Brown (11), Engberding (20), Hutchinson (46), and Miller (74). Again the evidence of Brown (11), Hutchinson (41), MacLennan (47, 48), Kelly (52), Koch (54), Krueger (57), Lipman (62, 63), Lipman, Brown and Owen (66, 67), Paterson and Scott (83), and Peck (87), points to the fact that lime increases ammonification. The stimulating effect of lime upon nitrification has been investigated by Brown (11), Densch (17), Fischer (24), Fittbogen (26), Fraps (28), Fred (52), Kelley (51, 52), Lyon and Bizzell (67), Lipman (62), McBeth and Wright (71), Miller (73), Müller and Weiss (79), Owen (82), Peck (86), Scales (96), Wilkins (120), and Wright (123). Denitrification studies have been performed by Lemmerman and Fischer (60), Lipman (62),

⁴ Burgess has found that nitrification is increased by fine material compared with coarse. Burgess, P. S., Nitrification as a measure of the availability of different forms of calcium carbonate when employed as correctors of soil acidity. *In* Soil Sci. (soon to appear).

and Vogel (112). Finally, the influence of lime on nitrogen-fixation has been investigated by Brown (11), Engberding (20), Hoffman and Hammer (42), Koch (55), Krainskii (56), Krueger (57), Lipman (62), and Vogel (112).

In the light of the interesting differences obtained in crop yield and neutralization of soil acidity it seemed advisable to carry out some experiments to determine the effect of different degrees of fineness of pulverized limestone on ammonification, nitrification and nitrogen-fixation. An attempt was made to correlate these biological activities with the rate of neutralization of soil acidity in the following manner. Norfolk sandy loam and Penn clay loam were incubated for seven weeks in 5-pound glazed earthenware pots with different grades of pulverized limestone and burnt lime, under the same conditions as described in the previous experiments. Ten cubic centimeters of an infusion made from soil known to contain active ammonifying and nitrifying bacteria; as well as 10 cc. of an infusion made from "Alphano humus," known to contain active nitrogen-fixing organisms, were added to each pot of soil. At the end of three weeks, when the soils were first sampled to determine the rate of neutralization of acidity, 5-grams portions of moist soil were immediately introduced into sterile flasks of media for the measurement of biological activities. These were prepared as follows, according to the methods described by Löhnis and Green (66).

A soil extract was prepared from each soil by adding 2 volumes of distilled water to 1 volume of soil and autoclaving in cotton-plugged Erlenmeyer flasks at 15 pounds pressure for fifteen minutes. After cooling and settling the supernatant liquid was filtered through several thicknesses of filter paper until a clear extract was obtained. To this was added 0.05 gram of K_2HPO_4 per litre. For ammonification, 50 cc. of this solution to which had been added 0.5 gram of peptone per flask were inoculated (in a 750-cc. Florence flask) with a 5-gram sample of soil and incubated at 23°C. for three days. At the end of this time the contents of the flasks were distilled with magnesium oxide and titrated in the usual manner. For nitrification, to 50 cc. of the extract of the soil type used as inoculum were added 50 mgm. of ammonium sulfate and 1 gram of calcium carbonate (c.p.). These flasks were incubated after inoculation with 5 grams of soil for four weeks. The nitrates were determined colorimetrically by the phenoldisulphonic acid method. For nitrogen-fixation 2 grams of dextrose and 2 grams of calcium carbonate were added to 50 cc. of soil extract and incubated for three weeks, at the end of which time they were Kjeldahled. Suitable checks were carried in duplicate in all instances and all the determinations on each soil were made in duplicate. Since the soils received duplicate treatments of lime the possibilities for error were reduced to a minimum.

The results of the ammonification of peptone after three weeks' incubation of soil with lime are recorded in table 7.

It will be seen that in both the Norfolk sandy loam and the Penn clay loam (with one exception) there is an increase in ammonification with an increase in fineness of division of pulverized limestone. It is of interest to note that while the differences in ammonification resulting from the finer grades of limestone are comparatively slight, there is a striking difference between the coarse and the finer limestone in both soils. In the Norfolk sandy loam,

for example, there was an increase of only 0.1 mgm. of nitrogen over the check for the 20-mesh as compared with 7.8 mgm. of nitrogen for the 200-mesh limestone. While in the Penn clay loam it was 0.3 mgm. of nitrogen for the 20-mesh as against 3.3 mgm. of nitrogen for the 200-mesh limestone. The burnt lime was more effective than the finest limestone in its influence on this biological activity. As might be expected from the more advantageous environmental conditions, the Norfolk sandy loam was responsible for greater differences between treatments than was the Penn clay loam.

Regarding these results in their entirety, it will be remembered that they are in accord with those obtained in determining the rate of neutralization of the different grades of pulverized limestone.

TABLE 7

The effect of fineness of division of pulverized limestone on the ammonification of peptone in solution (soil incubated with lime three weeks)

TREATMENT	NITROGEN mgm.	NITROGEN mgm.	AVERAGE NITROGEN mgm.	INCREASE IN NITROGEN mgm.
<i>Norfolk sandy soil</i>				
Check.....	26.0	26.1	26.1	
20- 40 mesh.....	26.2	Lost	26.2	0.1
60- 80 mesh.....	31.8	31.7	31.8	5.7
100-200 mesh.....	32.9	32.6	32.8	6.7
200 + mesh.....	32.0	35.7	33.9	7.8
CaO.....	35.6	37.3	36.5	10.4
<i>Penn clay loam</i>				
Check.....	23.0	26.0	24.6	
20- 40 mesh.....	23.6	26.1	24.9	0.3
60- 80 mesh.....	27.4	27.4	27.4	2.8
100-200 mesh.....	25.0	28.6	26.9	2.3
200 + mesh.....	27.1	28.4	27.9	3.3
CaO.....	29.7	29.0	29.4	4.8

The results at the end of seven weeks of incubation of soil with limestone are presented in table 8. It is evident that the differences between treatments in both soils are hardly large enough to bear mention, except in so far as the 20-mesh limestone gave the highest ammonification. This again may be correlated very closely with the results obtained concerning the rate of neutralization of acidity. In other words, the finer the limestone the more rapidly it neutralizes the acidity of the soil and the more effectively it increases the process of ammonification. It will be remembered that the 20-mesh material was the only one to be found present to any extent at the end of the seven-week period; therefore, it might well be expected that the present maximum of ammonia should occur with that grade of limestone.

Because of the importance of soil reaction in influencing nitrification, one would be led to anticipate marked differences in the accumulation of nitrates as a result of treating the soil with different grades of pulverized limestone. That this is virtually the case is shown by table 9, where it will be seen that with such widely divergent soil types, the differences in treatment at the end of three weeks yield results which are in identically the same direction, namely, an increase in fineness of division of pulverized limestone is responsible for an increased accumulation of nitrates. Again the burnt lime proved superior and the 200-mesh is strikingly better than the 20-mesh limestone in influencing this important biological activity in the soil. Thus a correlation exists between the rate of neutralization and nitrification as well as ammonifica-

TABLE 8

The effect of fineness of division of pulverized limestone on the ammonification of peptone in solution (soil incubated with lime seven weeks)

TREATMENT	NITROGEN mgm.	NITROGEN mgm.	AVERAGE NITROGEN mgm.
<i>Norfolk sandy loam</i>			
Check.....	42.8	43.3	43.1
20- 40 mesh.....	43.6	46.2	44.9
60- 80 mesh.....	41.0	40.8	40.9
100-200 mesh.....	41.1	Lost	41.1
200 + mesh.....	42.8	41.7	42.3
CaO.....	41.8	43.5	42.2
<i>Penn clay loam</i>			
Check.....	32.0	33.2	32.6
20- 40 mesh.....	33.5	32.6	33.1
60- 80 mesh.....	32.8	33.3	33.1
100-200 mesh.....	33.0	31.0	32.0
200 + mesh.....	32.4	33.1	32.8
CaO.....	31.6	32.9	32.3

tion. Coleman⁶ has a comprehensive discussion and bibliography dealing with nitrogen transformations in the soil. Since the accompanying data pointed indubitably to the fact that one could expect little, if any, differences in nitrification after seven weeks' incubation of soil with limestone, these determinations were discontinued.

The results obtained in the nitrogen-fixation experiments at the end of three weeks have not been tabulated for the reason that no differences worthy of note were established. For example, in the Norfolk sandy loam at the end of three weeks, the average number of milligrams of nitrogen present in the check soils was 3.37, while the amount in the variously treated soils ranged from the

⁶ Doctor's Thesis, Rutgers College, New Brunswick, N. J. (1917).

above-mentioned quantity up to 3.71 mgm. of nitrogen. In the Penn clay loam the check had 4.63 mgm. of nitrogen, and the range in variation due to treatment approached 5.04 mgm. of nitrogen as a maximum. At the end of seven weeks there was a distinct increase over the check in the soils treated with limestone, as shown in table 10, but the differences between treatments were not within experimental error in the case of Norfolk sandy loam. Thus 3.8 mgm. of nitrogen were present in the check soils and 12.3 to 12.9 mgm. of nitrogen in the treated soils. In Penn clay loam, however, there is an apparent tendency for the amount of nitrogen fixed to increase as the fineness of division of pulverized limestone increased.

TABLE 9

The effect of fineness of division of pulverized limestone on nitrification in solution (soil incubated with limestone three weeks)

TREATMENT	NO ₂	NO ₃	AVERAGE	INCREASE OVER CHECK
<i>Norfolk sandy loam</i>				
Check.....	7.4	6.8	7.1	
20- 40 mesh.....	7.9	8.2	7.8	0.7
60- 80 mesh.....	8.2	8.2	8.2	1.1
100-200 mesh.....	9.2	9.8	9.5	2.4
200 + mesh.....	10.2	13.0	11.6	4.5
CaO.....	9.4	20.0	14.7	7.6
<i>Penn clay loam</i>				
Check.....	7.6	8.4	8.0	
20- 40 mesh.....	11.4	18.4	14.9	6.9
60- 80 mesh.....	15.4	15.2	15.3	7.3
100-200 mesh.....	12.6	22.8	17.7	9.7
200 + mesh.....	20.6	22.0	21.3	13.3
CaO.....	24.0	28.0	26.0	18.0

Considering this experiment in its entirety, it has been established that both in Norfolk sandy loam and in Penn clay loam an increase in fineness of division of pulverized limestone was responsible for a corresponding increase in the rate of neutralization of acidity as well as enhanced biological activities in the soil as measured by ammonification, nitrification and nitrogen-fixation (with limitations) in solution.

In order to substantiate these points a similar experiment was performed in which Sassafras sandy loam was used in the same manner as previously described in the experiment dealing with the rate of neutralization of acidity, that is, burnt lime and the following grades of limestone were used: 20 to 40-mesh, 60 to 80-mesh, 100 to 200-mesh and finer than 200-mesh. In the present case, however, the fresh soil methods recommended by Brown (12, 13) were

employed. For ammonification, 100 grams of moist soil were thoroughly mixed in a shaker (61) with cottonseed meal or dried blood containing 155 mgm. of nitrogen, respectively, and incubated at optimum moisture content for six days at 20°C., the ammonia being determined by the magnesium oxide method at the end of this time. For nitrification 100 mgm. of ammonium sulfate were added per tumbler and incubated at optimum moisture content for thirty days at 18°C. At the end of this time the nitrates were leached out by adding 400 cc. of distilled water, some ground burnt lime was added and the whole shaken thoroughly for five minutes, filtered, and a 25-cc. aliquot taken for determination according to the colorimetric phenoldisulphonic acid method. For nitrogen-fixation 2 grams of mannite were added to 50

TABLE 10

The effect of fineness of division of pulverized limestone on the rate of neutralization of acidity as affecting nitrogen fixation in solution after seven weeks incubation of soil with limestone

TREATMENT	NITROGEN	NITROGEN	AVERAGE NITROGEN
<i>Norfolk sandy loam</i>			
Check.....	3.8	3.8	3.8
20- 40 mesh.....	11.4	13.2	12.3
60- 80 mesh.....	12.1	12.4	12.3
100-200 mesh.....	12.5	Lost	12.5
200 + mesh.....	12.6	13.2	12.9
CaO.....	11.5	13.3	12.4
<i>Penn clay loam</i>			
Check.....	4.1	5.4	4.8
20- 40 mesh.....	5.2	6.4	5.8
60- 80 mesh.....	7.0	8.5	7.8
100-200 mesh.....	7.5	8.0	7.8
200 + mesh.....	7.6	8.7	8.2
CaO.....	8.0	8.4	8.2

grams of soil and incubated at optimum moisture content for twenty-one days at 18°C. At the end of this period the soil was air-dried, pulverized and nitrogen determined by the Kjeldahl method.

The results of ammonification after one week's incubation of soil with limestone are recorded in table 11.

It will be seen that with cottonseed meal an increase in fineness of division of pulverized limestone was responsible for a corresponding increase in ammonia found. The 200-mesh is one-third more effective than the 20-mesh limestone, but burnt lime is somewhat superior to the former. The results obtained with dried blood are peculiar in that the 20- and the 60-mesh limestone yielded smaller quantities than were found in the checks, while the

finer grades of limestone and burnt lime exhibited an increase. This point will bear more complete discussion in another connection.

With cottonseed meal the results in table 11 obtained after two weeks' incubation (with one exception) show that an increase in fineness of division of pulverized limestone is accompanied by an increase in ammonification. However, the differences between treatments are not quite as pronounced as after the first week, undoubtedly because of the fact that the different grades of limestone tend to equalize, as is shown by the work on the rate of neutralization of acidity. Again the 200-mesh is markedly superior to the 20-mesh

TABLE 11

The effect of fineness of division of pulverized limestone on the ammonification of cottonseed meal and dried blood (Sassafras sandy loam incubated with lime for one and two weeks)

TREATMENT	COTTONSEED MEAL			DRIED BLOOD		
	Nitrogen	Nitrogen	Average nitrogen	Nitrogen	Nitrogen	Average nitrogen
<i>After one week's incubation</i>						
Check.....	47.1	49.3	48.2	42.5	43.5	43.0
20- 40 mesh.....	50.0	50.5	50.3	37.6	38.3	38.0
2.5 × 20-40 mesh.....	47.8	51.5	49.7	38.3	38.9	38.6
60- 80 mesh.....	60.4	56.4	58.4	32.8	35.2	34.0
100-200 mesh.....	53.7	66.5	60.1	41.3	45.7	43.5
200 + mesh.....	65.8	68.0	66.9	48.5	51.7	50.1
CaO.....	68.3	68.9	68.6	46.9	53.6	50.2
<i>After two weeks' incubation</i>						
Check.....	53.1	54.7	53.9	56.6	53.3	55.0
20- 40 mesh.....	55.7	54.7	55.2	47.2	46.7	47.0
2.5 × 20-40 mesh.....	57.6	57.0	57.3	43.5	41.8	42.7
60- 80 mesh.....	64.0	64.6	64.3	33.0	37.9	35.5
100-200 mesh.....	63.7	61.0	62.4	36.0	26.6	31.3
200 + mesh.....	65.0	64.6	64.8	27.4	27.5	27.5
CaO.....	65.6	66.6	66.1	30.4	28.6	29.5

limestone but inferior to the burnt lime. A point of interest is to be remarked in the fact that two and one-half times the usual quantity of 20-mesh limestone does not increase ammonification to any marked degree over that obtained with the normal amount, and is decidedly inferior to the 200-mesh limestone. The results with dried blood show a progressive decrease in ammonia with an increase in fineness of division of pulverized limestone. The explanation which suggests itself is that the ammonification of dried blood is actually more rapid with an increase in fineness of division of pulverized limestone but that the bacteria multiply so rapidly as to assimilate it again, so that it does not appear as ammonia when distilled with MgO, at the end of the six-day

incubation period, but is transformed into other nitrogenous compounds. This theory is substantiated by further experiments where the writer has found an increase in ammonification of dried blood with an increase in fineness of division of pulverized limestone. Furthermore, preliminary experiments were performed in which the ammonification of dried blood was accompanied by a daily measurement of carbon dioxide evolved. This was accomplished by aspirating air continuously through a stoppered bottle containing soil, which was connected with an Erlenmeyer flask containing a known quantity of standard barium hydrate. It was found that with an increase in fineness of division of pulverized limestone there was an increased production of carbon dioxide which may well be regarded as an index of decomposition effected by microbial activities. Thus it may be assumed that an increase in fineness of division of pulverized limestone actually caused an increase in the ammonification of dried blood, but that because of a subsequent assimilation of ammonia the amounts of ammonia found by distilling with magnesium oxide were comparatively small.

TABLE 12

The effect of fineness of division of pulverized limestone on ammonification (Sassafras sandy loam incubated with lime for three weeks)

TREATMENT	NITROGEN	NITROGEN	AVERAGE NITROGEN
<i>Cottonseed meal</i>			
Check.....	50.3	47.5	48.9
20- 40 mesh.....	49.4	57.1	53.3
2.5 X 20-40 mesh.....	52.6	54.1	53.4
60- 80 mesh.....	62.6	63.2	62.9
100-200 mesh.....	63.0	50.1	56.6
200 + mesh.....	59.0	56.0	57.5
CaO.....	48.2	56.2	52.2
<i>Dried blood</i>			
Check.....	52.8	48.5	50.7
20- 40 mesh.....	41.6	44.5	43.1
2.5 X 20-40 mesh.....	41.9	40.0	41.0
60- 80 mesh.....	28.6	35.0	31.8
100-200 mesh.....	31.0	20.0	25.5
200 + mesh.....	22.2	22.4	22.3
CaO.....	20.6	21.6	21.1

In tables 12 and 13 are recorded the results obtained after incubating soil with limestone for three and four weeks, respectively. With cottonseed meal as well as dried blood the general tendencies heretofore noted are again to be observed.

Considering the results obtained in the nitrification of ammonium sulfate after one week's incubation of soil with lime, it will be seen from table 14,

TABLE 13

The effect of fineness of division of pulverized limestone on ammonification (Sassafras sandy loam incubated with limestone four weeks)

TREATMENT	NITROGEN	NITROGEN	AVERAGE NITROGEN
<i>Cottonseed meal</i>			
Check.....	35.4	35.2	35.3
20- 60 mesh.....	32.5	37.0	34.8
2.5 × 20-60 mesh.....	37.4	39.0	38.2
60- 80 mesh.....	41.0	38.2	39.6
100-200 mesh.....	40.3	37.3	38.8
200 + mesh.....	42.1	38.9	40.5
CaO.....	33.1	36.6	33.9
<i>Dried blood</i>			
Check.....	32.3	32.5	32.4
20- 60 mesh.....	28.2	29.3	28.8
2.5 × 20-60 mesh.....	26.7	28.2	27.5
60- 80 mesh.....	24.0	24.6	24.3
100-200 mesh.....	21.8	14.9	18.4
200 + mesh.....	17.9	17.0	17.5
CaO.....	13.9	15.2	14.6

TABLE 14

The effect of fineness of division of pulverized limestone on nitrification (Sassafras sandy loam incubated with limestone one and two weeks)

TREATMENT	NITROGEN	NITROGEN	AVERAGE NITROGEN	INCREASE OVER CHECK
<i>After one week's incubation</i>				
Check on soil.....	0.06	0.06	0.06	
Check.....	0.13	0.12	0.13	0.07
20- 40 mesh.....	0.12	0.17	0.15	0.09
2.5 × 20-40 mesh.....	0.43	0.46	0.45	0.39
60- 80 mesh.....	0.51	0.96	0.74	0.68
100-200 mesh.....	1.28	0.70	0.99	0.93
200 + mesh.....	Lost	Lost	Lost	Lost
CaO.....	0.55	0.23	0.39	0.33
<i>After two weeks' incubation</i>				
Check.....	0.10	0.09	0.10	0.04
20- 40 mesh.....	0.32	0.17	0.25	0.19
2.5 × 20-40 mesh.....	2.46	1.48	1.97	1.37
60- 80 mesh.....	4.00	4.90	4.45	4.39
100-200 mesh.....	1.48	0.31	0.90	0.84
200 + mesh.....	0.31	0.65	0.48	0.42
CaO.....	0.15	0.11	0.13	0.07

that an increase in fineness of division of pulverized limestone is responsible for a corresponding accumulation of nitrates. The results obtained in the subsequent weeks of incubation, as shown in tables 14 and 15—are more variable, because the moisture conditions were not strictly maintained. However, the tendency follows the general rule previously observed, namely, an increase in fineness of division of pulverized limestone is responsible for an increase in biological activities, which may be correlated with the rate of neutralization of acidity. In the present experiment the differences in nitrogen-fixation, as a result of variation in treatment, were not of sufficient magnitude to permit of any definite conclusions. The average number of milligrams of nitrogen found varied from 0.98 to 1.21. It is possible that the amount of carbohydrate used in the present instance was so excessive as to inhibit nitrogen-fixation.

In order to emphasize further the importance of fineness of division of pulverized limestone in influencing ammonification, several soils (the same

TABLE 15
The effect of fineness of division of pulverized limestone on nitrification (Sassafras sandy loam incubated with lime for three weeks)

TREATMENT	NITROGEN mgm.	NITROGEN mgm.	AVERAGE NITROGEN mgm.	INCREASE OVER CHECK mgm.
Check.....	0.46	0.43	0.30	0.24
20- 40 mesh.....	0.37	0.17	0.27	0.21
2.5 X 20-40 mesh.....	1.96	1.95	1.95	1.90
60- 80 mesh.....	3.01	2.71	2.86	2.80
100-200 mesh.....	Lost	0.94	0.94	0.88
200 + mesh.....	1.20	1.37	1.29	1.23
CaO.....	0.75	0.72	0.74	0.68

as previously used in growing clover in pots) were incubated with the different grades of limestone and burnt lime for one week. At the end of this time 100-gram portions from each pot received nitrogenous organic material in the form of dried blood or cottonseed meal (155 mgm. N), and were incubated at the optimum moisture content for seven days at 20°C. The results obtained with Cumberland silt loam are recorded in table 16.

Although the differences between treatments are not great, nevertheless it will be seen that in general, with both cottonseed meal and dried blood, an increase in fineness of division of pulverized limestone is accompanied by an increase in ammonia found. The burnt lime and 200-mesh limestone were equally efficient. Two and one-half times the usual quantity of 20-mesh limestone gives better results, not markedly superior to the normal application.

The results obtained with Wooster and Carrington silt loams are presented in table 16. The differences between treatments are not very marked but the finer material appears to better advantage than the coarser. Burnt lime

proved more efficient than 200-mesh limestone. The results obtained with increased quantities of 20-mesh limestone are similar to those noted where Cumberland silt loam was employed.

A general consideration of these soils of fine texture and high organic content reveals the fact that in the ammonification of nitrogenous organic ma-

TABLE 16

The effect of fineness of division of pulverized limestone on ammonification in Cumberland, Carrington and Wooster silt loams

TREATMENT	COTTONSEED MEAL			DRIED BLOOD		
	Nitrogen	Nitrogen	Average nitrogen	Nitrogen	Nitrogen	Average nitrogen
<i>Cumberland silt loam</i>						
Check.....	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
Check.....	70.0	70.7	70.4	78.3	77.4	77.9
20- 40 mesh.....	74.9	69.3	72.1	64.8	66.6	65.7
2.5 X 20-40 mesh.....	Lost	73.8	73.8	67.8	64.8	66.3
60- 80 mesh.....	73.4	72.0	72.7	61.4	71.8	66.6
100-200 mesh.....	73.8	74.3	74.1	59.8	62.5	61.2
200 + mesh.....	75.2	74.5	74.9	81.9	85.5	83.7
CaO.....	74.5	72.0	73.3	82.6	82.3	82.5
<i>Wooster silt loam</i>						
Check.....	40.1	43.4	41.8	50.2	50.0	50.1
20- 40 mesh.....	47.0	49.7	48.4	50.0	47.7	48.9
2.5 X 20-40 mesh.....	50.4	50.2	50.3	49.3	49.1	49.2
60- 80 mesh.....	40.5	41.5	41.0	45.2	45.5	45.4
100-200 mesh.....	49.1	50.0	49.6	42.8	43.0	42.9
200 + mesh.....	48.8	49.3	49.1	58.7	59.4	59.1
CaO.....	59.4	58.3	58.9	69.3	63.4	66.4
<i>Carrington silt loam</i>						
Check.....	33.8	41.0	37.4	58.1	57.4	57.8
20- 40 mesh.....	37.3	50.0	43.7	59.9	61.6	60.3
2.5 X 20-40 mesh.....	57.8	27.7	42.8	60.8	61.6	61.2
60- 80 mesh.....	38.3	47.5	42.9	53.6	53.3	53.5
100-200 mesh.....	47.7	46.3	47.0	65.7	61.7	63.7
200 + mesh.....	Lost	Lost	Lost	53.2	60.0	61.6
CaO.....	50.2	50.2	50.2	66.6	Lost	66.6

terials the differences resulting from treatment with various grades of limestone are less marked than those noted where soils of coarser texture and lower organic content were employed. However, the evidence points quite definitely to the contention that the finer grades of limestone exert a more beneficial influence upon the process of ammonification than does the coarse material. This is especially significant for the light it throws upon certain pre-

vious results obtained in the ammonification of dried blood and would seem to make the theory advanced in that connection more tenable.

Since in common field practice the amount of limestone applied to the soil is rarely the exact equivalent of the lime-requirements as determined by the

TABLE 17

The effect of fineness of division of pulverized limestone in varying amounts and CaO on ammonification in Norfolk sandy loam. Cottonseed meal

TREATMENT	NITROGEN	NITROGEN	AVERAGE NITROGEN
<i>One-third Veitch lime-requirement applied</i>			
Check.....	50.0	51.6	50.8
0.09 gram, 20- 40 mesh.....	56.5	54.7	55.6
0.09 gram, 60- 80 mesh.....	56.1	59.2	57.7
0.09 gram, 100-200 mesh.....	61.6	61.7	61.1
0.09 gram, 200 + mesh.....	60.5	60.6	60.6
0.05 gram, CaO.....	54.2	54.5	54.4
<i>Two-thirds Veitch lime-requirement applied</i>			
Check.....	50.0	51.6	50.8
0.18 gram, 20- 40 mesh.....	54.1	54.9	54.4
0.18 gram, 60- 80 mesh.....	52.8	53.8	53.3
0.18 gram, 100-200 mesh.....	56.8	52.9	54.9
0.18 gram, 200 + mesh.....	56.3	55.9	56.1
0.10 gram, CaO.....	65.3	63.1	64.2
<i>Total Veitch lime-requirement applied</i>			
Check.....	50.0	51.6	50.8
0.28 gram, 20- 40 mesh.....	51.5	50.9	51.2
0.28 gram, 60- 80 mesh.....	55.3	54.9	55.1
0.28 gram, 100-200 mesh.....	59.2	59.8	59.5
0.28 gram, 200 + mesh.....	62.8	63.1	63.0
0.15 gram, CaO.....	65.2	65.2	65.2
<i>One and one-third Veitch lime-requirement applied</i>			
Check.....	50.0	51.6	50.8
0.37 gram, 20- 40 mesh.....	57.7	59.0	58.4
0.37 gram, 60- 80 mesh.....	63.6	63.6	63.6
0.37 gram, 100-200 mesh.....	58.8	58.6	58.7
0.37 gram, 200 + mesh.....	65.8	69.0	67.4
0.20 gram, CaO.....	68.0	69.7	68.9

Veitch method, the following experiment was performed. Limestone of different grades of fineness and burnt lime were applied to Norfolk sandy loam at the rate of one-third, two-thirds, total and one and one-third of the Veitch lime-requirement of 2300 pounds of CaO per acre. One series received 155

mgm. of nitrogen in the form of cottonseed meal, the other received dried blood. The soils were made up to the optimum moisture content (13 per cent) and incubated for seven days, the ammonia being determined by the magnesian oxide method at the end of that time. The results are presented in tables 17 and 18.

TABLE 18

The effect of fineness of division of pulverized limestone in varying amounts and CaO on ammonification in Norfolk sandy loam (dried blood being used)

TREATMENT	NITROGEN mgm.	NITROGEN mgm.	AVERAGE NITROGEN mgm.
<i>One-third Veitch lime-requirement applied</i>			
Check.....	70.6	69.8	70.2
0.09 gram, 20- 40 mesh.....	71.6	71.6	71.6
0.09 gram, 60- 80 mesh.....	69.5	69.6	69.6
0.09 gram, 100-200 mesh.....	65.4	67.0	66.2
0.09 gram, 200 + mesh.....	59.5	60.7	66.2
0.05 gram, CaO.....	67.7	67.4	67.6
<i>Two-thirds Veitch lime-requirement applied</i>			
0.18 gram, 20- 40 mesh.....	67.3	66.5	66.9
0.18 gram, 60- 80 mesh.....	64.2	65.0	64.6
0.18 gram, 100-200 mesh.....	60.7	60.2	60.5
0.18 gram, 200 + mesh.....	58.4	59.5	59.0
0.10 gram, CaO.....	61.1	59.0	60.1
<i>Total Veitch lime-requirement applied</i>			
0.28 gram, 20- 40 mesh.....	64.9	65.8	65.4
0.28 gram, 60- 80 mesh.....	64.3	62.2	63.3
0.26 gram, 100-200 mesh.....	59.5	59.1	59.3
0.26 gram, 200 + mesh.....	60.1	60.2	60.2
0.15 gram, CaO.....	59.3	59.9	59.6
<i>One and one-third Veitch lime-requirement applied</i>			
0.37 gram, 20- 40 mesh.....	67.9	70.1	69.5
0.37 gram, 60- 80 mesh.....	66.3	64.7	65.5
0.37 gram, 100-200 mesh.....	59.1	61.1	60.1
0.37 gram, 200 + mesh.....	60.6	59.3	60.0
0.20 gram, CaO.....	63.2	63.7	63.5

It will be seen that with but a few exceptions in the cottonseed meal series, an increase in fineness of division of pulverized limestone is responsible for an increased ammonification. This phenomenon appeared regardless of the size of application of limestone. An increase in the quantity of burnt lime caused a still further increase in ammonification. In the dried blood series an increase in fineness of division of limestone caused a corresponding division

in ammonia found. However, as previously interpreted, this may be considered as virtually representing an increase in ammonification accompanied by an increase in assimilation with an increase in fineness of division of pulverized limestone. Again the phenomenon remains constant, regardless of the rate of application of limestone. In general, then, it appears that ammonification as influenced by the different grades of pulverized limestone is independent of the size of application.

Summarizing the results obtained in the present experiments, the following points are worthy of note.

1. An increase in fineness of division of pulverized limestone was responsible for an increase in the ammonification of peptone in solution in Norfolk sandy loam and Penn clay loam, and of cottonseed meal and dried blood in Sassafras sandy loam, which may be correlated with the results obtained in the rate of neutralization of acidity. That is, in the first two weeks of incubation of soil with limestone the differences between treatments are decidedly more marked than in the following weeks when there is a general equalizing tendency manifest.
2. An increase in fineness of division of pulverized limestone is responsible for an increase in nitrification and nitrogen-fixation in solution and fresh soil, which may also be correlated with the rate of ammonification.
3. The ammonification of dried blood and cottonseed meal followed the above tendency in the heavy soils rich in organic matter as well as the lighter soils used.
4. These results of ammonification remain constant regardless of the size of the application of limestone (as determined by the Veitch method).

THE INFLUENCE OF FINENESS OF DIVISION OF PULVERIZED LIMESTONE ON LOSS BY DRAINAGE⁶

So far as the writer has been able to determine, the recent publication from the Maryland Agricultural Experiment Station (12) represents the sole work on the solubility and loss of different degrees of pulverized limestone when applied to the soil. In the experiments performed by Broughton, Williams, and Frazer (10), both limestone and oyster shells of varying degrees of fineness were employed. The solubility of these samples which had been subjected to mechanical analysis was determined in water, and water charged with carbon dioxide at 3°, 12°, 14°, and 23°C., representing the temperatures of winter, spring, autumn, and summer, respectively. It was found that with an increase in fineness of division of pulverized limestone there was an increase in solubility, the same phenomenon being observed with an increase in temperature. The solubility and the different materials were then determined

*The writer wishes to express appreciation for his assistance to Mr. R. C. Cook, with whom this experiment was begun. Owing to unforeseen circumstances the latter was unable to continue this work.

by analyzing the soil solution obtained from large pots treated with lime. Unfortunately, no duplicate treatments are mentioned, and since the soils considered as checks received applications of either CaO or CaCO₃, the experiment is open to criticism on these grounds. This is further emphasized by the fact that the differences between treatments were slight. However, it was shown that the solubility of these materials increased with an increase in the fineness of division of limestone. In other words the greater the surface of limestone exposed, the greater was its solubility. Two definite conclusions were recorded from interesting data of field experiments still in progress. The authors maintain that in order to furnish as much calcium as is present in burnt lime, limestone should be ground so that 90 per cent passes an 80-mesh sieve. Their conclusion is corroborated by the work reported in this thesis.

There is a considerable amount of data concerning the distribution, changes and loss of limestone by leaching which has been reported by Blanck (7), Broughton (9), Creydt, Von Seelhorst and Wiems (16), Frear (30), Hilgard (40), Johnson (49), Lyon and Bizzell (69), MacIntyre, et al. (72), Miller (74), and Veitch (109). The effect of adding limestone in excess of the amount required to neutralize acidity has been studied by Gardner and Brown (34), White (107), and Veitch (109).

In order to study the influence of the fineness of pulverized limestone on the loss of lime in drainage, the following method was devised and put into operation in 1914.⁷

Large galvanized iron pots, 16 inches in diameter and 12 inches deep, standing on three legs, were fitted with stopcocks to permit drainage. The bottom of the pot had a slight depression in which were placed large pebbles. The pots were fitted with a pair of handles riveted to the sides, and the whole painted to prevent rusting. A layer of fine pebbles was placed in the bottom of the pot, and over this was placed 120 pounds of Sassafras sandy loam which had been sieved through an 8-mesh screen (plate 1). A lever arrangement with grippers for the handles of the pot, and a counterpoise was devised for keeping the pots at constant weight. Two series of new pots were placed upon a greenhouse table. A hole was drilled under each stopcock to permit the fastening of a rubber tube which extended down into a bottle to collect the drainage water. A board support was nailed close to the ground upon which the bottles might be placed in proper position. After the harvesting of each crop, the drainage water was collected by flushing through enough water to obtain 3800 cc. of drainage in each collecting bottle. This method is, of course, open to objection, in that continuous percolation would give a more adequate representation of the drainage water, but the greenhouse conditions were such as to make such a procedure impracticable.

⁷ Broughton (10) and the United States Bureau of Plant Industry, at Washington, D. C., have since described similar apparatus for the same purpose (122).

The experiment was continued over a period of time which permitted the growth of several different crops, the soil being treated with pulverized limestone of different grades in quantity sufficient to fulfill the lime-requirement as determined by the Veitch method. One series of pots received ammonium sulfate at the rate of 600 pounds to the acre, while the other series received no addition of nitrogen. The check pots received minerals above, while the others received minerals in addition to the special treatment. Before planting the first crop the soil from each pot was removed to a depth of 6½ inches, while 15 grams of acid phosphate (14 per cent P₂O₅) and 7.5 grams of muriate of potash (50 per cent K₂O) were thoroughly incorporated together with 57 grams of pulverized limestone of different grades of fineness. The soil, was designated as Sassafras sandy loam which had been unlimed, and unfertilized

TABLE 19

The effect of fineness of division of pulverized limestone on the yield of barley grown on Sassafras sandy loam in drainage tanks with and without the addition of nitrogen

TREATMENT	WEIGHT OF CROP	DUPLICATE	AVERAGE	NITROGEN	NITROGEN	AVERAGE NITROGEN	TOTAL NITROGEN
Without the addition of nitrogen							
Check.....	45.5	36.5	41.0	1.18	0.89	1.04	0.43
20- 40 mesh...	79.0	80.5	79.7	0.83	0.96	0.90	0.71
60- 80 mesh...	95.0	88.5	91.7	0.86	0.86	0.86	0.79
100-200 mesh...	93.0	91.5	92.2	0.88	0.84	0.86	0.79
200 + mesh....	81.5	85.0	83.2	0.82	0.84	0.83	0.69
12 grams (NH ₄) ₂ SO ₄ applied							
Check.....	48.0	55.5	51.7	1.30	1.32	1.31	0.68
20- 40 mesh...	82.0	Lost	82.0	1.28	1.33	1.31	1.07
60- 80 mesh...	87.0	73.0	80.0	1.33	1.08	1.21	0.97
100-200 mesh...	74.0	92.0	83.0	1.33	1.22	1.28	1.06
200 + mesh....	78.0	67.5	72.7	1.32	Lost	1.32	0.96

and had a lime-requirement (Veitch) of 100 pounds CaO per acre. The pots were maintained at optimum moisture content. Seven grams of ammonium sulfate were applied to each pot of one series one week before planting 100 seeds of barley per pot. The other 5 grams were added after the seedlings had been thinned to 50 plants.

The results of yield and nitrogen content of the first crop grown in these tanks is presented in table 19.

Some discrepancies which appear are due to attacks from fungi which did not invalidate the general tendency exhibited, namely, with an increase in the fineness of division of pulverized limestone up to the 200-mesh product there was an increase in crop yield and total nitrogen in grams. This is to be seen both in the series receiving no nitrogen and that receiving 12 grams of

ammonium sulfate per pot. From the figures recorded in the columns marked "average per cent of nitrogen" it is apparent that the differences as a result of variation in treatment are so slight as to be negligible. Thus the total nitrogen expressed in grams (last column) varies almost as the crop yield. The question which is immediately suggested by these results is: why are the yields obtained with the finest limestone smaller than those where the coarser material was used? The explanation which appears most plausible is that in such a light open sandy soil the finer material is washed down below the root zone of the plant, therefore it does not neutralize the soil acidity or increase the available nitrogen supply of the plant to the same extent as does the coarser material which is actually less efficient. This hypothesis is strengthened by certain subsequent results concerned with the planting of alfalfa (which is one of the deepest-rooted of all crops) which seems to respond with a greater yield to the finer limestone. Likewise, the fact that more lime is lost in leaching from the soils receiving the finer material, as will be shown presently, also may be considered as corroborative evidence. A further consideration may be advanced, namely, it is possible that the finer material, being more active, effects a reversion of the acid phosphate sufficient to limit plant growth to some degree. However, in view of the large amount of acid phosphate applied, this does not appear tenable. Again the nitrogen in the soil may have been oxidized to nitrates more rapidly with soils receiving the finer limestone, and rapidly transformed into insoluble nitrogenous compounds before the plant could use them. As has been previously stated, after harvesting the crops (which had been allowed to grow to maturity), water was flushed through the soil and the 3800 cc. of drainage water collected. Duplicate aliquot samples of 5 cc. were taken for the determination of ammonia by the Nessler colorimetric method (98), and 25-cc. portions were taken for nitrate nitrogen determinations according to the phenoldisulphonic acid method. The 3740 cc. of drainage water were then evaporated to about 400 cc. After 10 cc. of hydrochloric acid (concentrated) were added, the solution was made up to 500 cc. and kept in a tightly-stoppered Jena flask until used for the determination of calcium by the standard permanganate titration method.

From the results recorded in table 20, it will be noted that in both series an increase in fineness of division of pulverized limestone was accompanied by a decrease in loss of ammonia in the drainage water. As was to be expected the series receiving ammonium sulfate lost more ammonia than the series receiving no nitrogen. The determination of nitrates and calcium in the drainage waters led to no definite conclusion because of lack in agreement between duplicate treatments, therefore the results were not tabulated.

After permitting the soil to dry out sufficiently to restore the optimum moisture condition, buckwheat was planted and the crop grown to maturity. In this case, no further applications of limestone were made, but the usual 12 grams of ammonium sulfate were applied to each pot of the series receiving nitrogen. The yield and nitrogen content of the crops are recorded in table 21.

In the series receiving no nitrogen there is an increase in crop yield with 60 to 80-mesh as compared with 20 to 40-mesh limestone. The material finer than 60-mesh caused a decline in yield which, however, remained superior

TABLE 20
Analysis of first drainage water for ammonia

TREATMENT	NH ₃ IN 3800 CENTIMETERS	DUPLICATE NH ₃	AVERAGE NH ₃	DECREASE OVER CHECK NH ₃
<i>No nitrogen applied</i>				
Check.....	0.532	0.760	0.646	
20- 40 mesh.....	0.026	0.026	0.026	0.619
60- 80 mesh.....	0.030	0.019	0.025	0.621
100-200 mesh.....	0.011	0.019	0.015	0.630
200 + mesh.....	0.019	0.000	0.010	0.636
<i>12 grams (NH₄)₂SO₄ applied</i>				
Check.....	1.140	1.133	1.235	
20- 40 mesh.....	0.570	0.950	0.760	0.475
60- 80 mesh.....	0.836	0.608	0.722	0.513
100-200 mesh.....	0.570	0.228	0.399	0.836
200 + mesh.....	0.285	Lost	0.285	0.950

TABLE 21
The effect of fineness of division of pulverized limestone on the yield of buckwheat in drainage tanks with and without applications of nitrogen

TREATMENT	WEIGHT OF CROP	DUPLICATES	AVERAGE	NITROGEN	NITROGEN	AVERAGE NITROGEN	TOTAL NITROGEN
<i>Without nitrogen</i>							
	grams	grams	grams	per cent	per cent	per cent	grams
Check.....	38.0	15.7	26.9	1.99	1.95	1.97	0.53
20- 40 mesh...	44.9	43.0	44.0	1.31	1.43	1.37	0.60
60- 80 mesh...	68.7	71.7	70.2	1.17	1.33	1.25	0.88
100-200 mesh...	58.0	53.0	55.5	1.22	1.29	1.26	0.70
200 + mesh....	54.8	50.7	52.8	1.56	1.53	1.55	0.82
<i>12 grams (NH₄)₂SO₄ applied</i>							
	grams	grams	grams	per cent	per cent	per cent	grams
Check.....	16.0	17.6	16.8	2.37	2.37	2.37	0.40
20- 40 mesh...	80.4	88.6	84.5	1.96	2.36	2.16	1.82
60- 80 mesh...	93.5	103.4	99.0	1.86	2.14	2.05	2.03
100-200 mesh...	90.9	84.0	87.5	2.20	2.14	2.17	1.90
200 + mesh.	88.5	94.9	92.7	2.12	2.12	2.12	1.97

to the 20-mesh limestone. There was very little variation in the percentage of nitrogen, although in this instance the 200-mesh limestone gave a considerably higher percentage than did the other grades of limestone. The reason

for the high nitrogen content in the checks is, of course, the relatively low yield, this phenomenon being of too common occurrence to require further comment. Again in general, the total nitrogen in grams is roughly proportional to the yields obtained. In the series receiving ammonium sulfate it is at once evident that the total yield of all pots (with the exception of the checks, due to higher acidity), is higher than in the case of the soils receiving no nitrogen. Again, the results with the different grades of limestone and ammonium sulfate exactly parallel those obtained where nitrogen was not added. The variations in nitrogen content are negligible. The fact that the 60-mesh gives a superior yield to that obtained with the finer limestone has already been discussed in connection with the previous crop. It is especially interesting to note that the yields where fine pulverized limestone is

TABLE 22

Neutralization of acidity by different degrees of fineness of pulverized limestone (Sassafras sandy loam after growing buckwheat)

TREATMENT	CaO PER ACRE	CaO PER ACRE	AVERAGE CaO PER ACRE
<i>No nitrogen applied</i>			
Check.....	pounds	pounds	pounds
Check.....	4,900	4,600	4,750
20- 40 mesh.....	3,100	3,100	3,100
60- 80 mesh.....	2,600	1,600	2,100
100-200 mesh.....	1,750	1,650	1,700
200 + mesh.....	1,650	1,650	1,650
<i>12 grams (NH₄)₂SO₄ applied</i>			
Check.....	5,200	4,500	4,850
20- 40 mesh.....	3,600	3,400	3,500
60- 80 mesh.....	3,600	2,500	3,050
100-200 mesh.....	2,250	3,300	2,375
200 + mesh.....	1,900	2,000	1,950

used without any addition of nitrogen are only slightly inferior to the yields obtained where coarse limestone was accompanied by an application of ammonium sulfate at the rate of 660 pounds per acre.

In order to cast further light upon the neutralization of soil acidity by the different grades of pulverized limestone, the lime-requirement of all soils was determined after harvesting the crops and before leaching. This was done by taking three cores of soil the entire depth of the pot, mixing them thoroughly and measuring out a representative sample of 200 grams, of which 10-gram portions were used for the Veitch determinations. The results obtained are shown in table 22 and may be considered of primary importance.

In the first place it is evident in both series that an increase in fineness of division is responsible for a diminution of the lime-requirement. Again,

it will be seen that in comparing the series receiving ammonium sulfate with the series receiving no nitrogen, the former had a uniformly higher lime-requirement than the latter, regardless of the grades of limestone used. While the agreement between duplicates is not in all cases what might be desired, nevertheless, the general trend is unmistakable. This, after a fashion, may be considered as bearing upon the discussion of crop yield, namely, the composite sample of soil receiving 200-mesh limestone has a lower lime-requirement than where the coarser material was used, but the fact that the plants could not take advantage of this indicates that much of the neutralization took place below the root zone. Consequently, the yields obtained with medium fine material are superior to those obtained with very fine limestone.

After harvesting the crops, the soil in each pot was leached as previously described and duplicate samples of 20 cc. were taken for the determination of acidity by titration with N/50 NaOH, phenolphthalein being used; and of alkalinity by titration with N/50 HCl, with methyl orange as an indicator. The general tendency was for a decrease in acidity with an increase in fineness of division of pulverized limestone, and for an increase in alkalinity. The series receiving nitrogen showed a higher acidity than where ammonium sulfate was applied. Thus it required 0.86 cc. of N/50 NaOH to neutralize the acidity where 200-mesh was used, as against 1.75 cc. where 20-mesh limestone was used in the series receiving no nitrogen, and 1.46 cc. as against 1.96 cc. in the series receiving ammonium sulfate. The other results were not tabulated because of lack of sufficient agreement between duplicates. This is likewise true of the determinations of ammonia, nitrates and calcium, although the general tendency exhibited was that an increase in fineness of division of pulverized limestone was responsible for a decrease in the loss of ammonia and nitrate nitrogen and an increase in the loss of calcium in the drainage waters.

The next crop to be grown in these pots was rape. This was selected because of the fact that the experiment in progress required that there be no addition of nitrogen to one series of soils, and rape is not regarded as a heavy feeder on nitrogen. Of course, the root residues of former crops furnished a source of nitrifiable material and this is undoubtedly the principal reason for the good yields obtained. Fifteen grams of acid phosphate and 7.5 grams of muriate of potash were added at the outset of the experiment and limestone was added to all soils in excess of the lime-requirement as determined after harvesting buckwheat (table 22). These applications were made on the basis of two-thirds in excess of the lime-requirement of the soils having the greatest acidity (the checks receiving one-fourth in excess in the form of 200-mesh limestone). The additions of fertilizer and lime were made two weeks before planting 20 rape seeds. The seedlings were thinned to 10 in number, and the additional application of 5 grams of ammonium sulfate made. The crop was grown to maturity but difficulty was experienced in combatting cabbage worms. This pest was finally exterminated, after creating considerable dam-

age, by sprinkling the leaves with powdered lead arsenate. The results of crop yield and nitrogen content are recorded in table 23.

While the differences between treatments are not pronounced, due to the fact that an excess of lime was applied, nevertheless, it is evident in both series that the 60 to 80-mesh limestone yielded superior results while the 200-mesh appeared to better advantage than did the 20-mesh limestone, which corroborates the conclusions based on the yields with the previous two crops. Again the percentage of nitrogen in both series is highest with the 200-mesh limestone, while the total nitrogen in grams parallels the crop yields. It will also be seen the series receiving ammonium sulfate gave higher yields and nitrogen content than the series receiving no nitrogen. However, the yields

TABLE 23

*The effect of fineness of division of pulverized limestone on the yield of rape in drainage tanks
(two-thirds in excess of lime-requirement)*

TREATMENT	WEIGHT OF CROP	DUPLICATES	AVERAGE	NITROGEN	NITROGEN	AVERAGE NITROGEN	TOTAL NITROGEN
<i>No nitrogen applied</i>							
Check.....	55.4	68.4	61.9	1.50	1.55	1.53	0.95
20- 40 mesh...	56.0	60.0	58.0	1.31	1.29	1.30	0.75
60- 80 mesh...	71.0	72.5	71.8	1.29	1.31	1.30	0.93
100-200 mesh...	72.2	64.2	68.2	1.28	1.40	1.34	0.91
200 + mesh....	66.2	69.2	67.7	1.42	1.35	1.39	0.94
<i>12 grams (NH₄)₂SO₄ applied</i>							
Check.....	78.7	70.5	74.6	2.63	2.85	2.79	2.08
20- 40 mesh...	77.7	72.0	74.9	1.89	1.81	1.85	1.39
60- 80 mesh...	95.0	81.2	88.1	1.81	1.87	1.84	1.62
100-200 mesh...	76.7	71.0	73.9	1.88	1.88	1.88	1.39
200 + mesh....	76.0	76.6	76.3	1.88	1.92	1.90	1.45

with the finer limestone without nitrogen were only slightly inferior to those obtained with the coarse limestone where ammonium sulfate had been added at the rate of 600 pounds to the acre. Thus one might argue that limestone was responsible for an increased supply of available nitrogen to the plant.

Following the previous procedure of determining the lime-requirement of all the soils before leaching, the results are presented in table 24.

It will be seen that in both series an increase in fineness of division of pulverized limestone was accompanied by a decrease in the lime-requirement. There is mutual corroboration in the fact that the check soils, as well as those regularly receiving 200-mesh limestone, were alkaline, for it will be remembered that the former received an excessive application of 200-mesh limestone. It is further interesting to note that the acidity developed in the soils receiving

60-mesh is only one-half (or less) of that developed in the soils receiving 20-mesh limestone.

The soils were leached in the usual manner and the acidity and alkalinity titrated as before. Again, the results indicate that with an increase in fineness of division of pulverized limestone there is a decrease in acidity and an increase in alkalinity. For example, in the series receiving no nitrogen, 20 cc. of drainage water from the soil treated with 20-mesh requires 3.28 cc. of N/50 NaOH to effect neutralization while 200-mesh requires only 2.53 cc. In the series receiving ammonium sulfate, the former treatment requires 3.93 cc. and the latter only 1.80 cc. The results of the ammonia and nitrate determinations were too variable to permit of definite conclusions, although there

TABLE 24

Neutralization of acidity by different degrees of fineness of division of pulverized limestone (Sassafras sandy loam after growing rape). Two-thirds in excess of lime requirement applied—200-mesh limestone applied to check

TREATMENT	CaO pounds	CaO pounds	AVERAGE CaO pounds
<i>No nitrogen applied</i>			
Check.....	Alkaline	Alkaline	Alkaline
20- 40 mesh.....	2,500	2,300	2,400
60- 80 mesh.....	1,300	1,200	1,250
100-200 mesh.....	Alkaline	Alkaline	Alkaline
200 + mesh.....	Alkaline	Alkaline	Alkaline
<i>12 grams (NH₄)₂SO₄ applied</i>			
Check.....	100	Alkaline	50
20- 40 mesh.....	3,500	2,900	3,200
60- 80 mesh.....	1,900	800	1,350
100-200 mesh.....	100	Alkaline	50
200 + mesh.....	Alkaline	Alkaline	Alkaline

appeared to be a tendency for an increase in fineness of division of pulverized limestone to cause a decrease in loss of ammonia and nitrates. Thus in the series receiving ammonium sulfate the 20-mesh treated soils contained 2.10 mgm. of nitrogen nitrates, while the soils treated with 200-mesh yielded only 0.46 mgm. of nitrogen. Again in the series receiving no nitrogen, with the 20-mesh there were 1.653 mgm. of nitrogen in the form of ammonia in the drainage compared with 0.798 mgm. of nitrogen in the case of the 200-mesh.

The results of the calcium determinations in the drainage water are presented in table 25.

It will be seen that in both series (with one exception) there is an increased loss of lime with an increase in fineness of division of pulverized limestone.

While the duplicates are not always in close agreement, nevertheless, the general tendency of the averages is pronounced.

Since rape grew so well under the conditions of its first trial, it was deemed advisable to use the same plant again. No lime or fertilizer was added other than the ammonium sulfate in the manner previously described. Twenty-five seeds were planted in each pot, and these were later thinned to 15 seedlings. An inspection of the results in table 26 shows slight differences as a result of treatment.

However, as usual the 60-mesh limestone is superior to the other grades. The yields on the soils receiving ammonium sulfate are distinctly superior to those receiving no nitrogen. Because of the nature of these results, differences in drainage waters were not to be anticipated, and consequently the soils were not leached.

TABLE 25
Analysis of drainage water: determination of calcium. Third series

TREATMENT	Ca	DUPPLICATES	AVERAGE
<i>No nitrogen applied</i>			
Check.....	mgm.	mgm.	mgm.
Check.....	78.3	73.0	75.7
20- 40 mesh.....	77.9	Lost	77.9
60- 80 mesh.....	71.7	70.0	70.9
100-200 mesh.....	81.8	80.5	81.2
200 + mesh.....	85.4	89.8	87.6
<i>12 grams (NH₄)₂SO₄ applied</i>			
Check.....	51.9	79.2	65.6
20- 40 mesh.....	69.5	112.2	90.9
60- 80 mesh.....	153.1	49.3	101.2
100-200 mesh.....	176.0	Lost	118.4
200 + mesh.....	176.0	92.0	134.0

Summarizing the results obtained in the work just discussed concerning the growth of various crops in large pots permitting the collection of drainage water and the analyses thereof, the following points have been established, a light sandy loam (Sassafras) being treated with different degrees of fineness of pulverized limestone, which had been cropped to barley, buckwheat, and rape (twice).

1. Both in the series receiving an application of ammonium sulfate at the rate of 660 pounds to the acre and that receiving no nitrogen, the 60-mesh limestone gave the highest yields. This is explained by the fact that the finer grades of limestone were washed down below the root zone where they became less effective in neutralizing soil acidity and making plant-food available. The 200-mesh gave higher yields than 20-mesh limestone.

2. The highest average per cent of nitrogen in the various crops was obtained with the 200-mesh limestone in the series with and without nitrogen.

3. The soils receiving an application of ammonium sulfate at the rate of 600 pounds to the acre yielded crops superior to those which received no nitrogen, when the limestone treatment remained the same.

4. The best yields with the finer limestone with and without nitrogen are only slightly inferior to the yields obtained with the coarsest limestone on soil receiving 660 pounds of ammonium sulfate to the acre. This suggests a possible value for finely-pulverized limestone, namely, the making available of nitrogen in the soil.

TABLE 26

The effect of fineness of division of pulverized limestone on the second successive crop of rape grown in drainage pots

TREATMENT	WEIGHT OF CROP	DUPLICATES	AVERAGE
<i>Without nitrogen</i>			
Check.....	grams	grams	grams
Check.....	17.7	20.9	19.3
20- 40 mesh.....	19.8	21.5	20.7
60- 80 mesh.....	24.2	23.6	23.9
100-200 mesh.....	20.8	17.8	19.3
200 + mesh.....	18.0	23.1	20.6
<i>12 grams (NH₄)₂SO₄ applied</i>			
Check.....	29.1	10.3*	29.1
20- 40 mesh.....	32.2	32.2	32.2
60- 80 mesh.....	33.0	32.1	32.6
100-200 mesh.....	31.5	31.5	31.5
200 + mesh.....	25.3*	31.6	31.6

* Poor growth observed.

5. Large galvanized iron pots were devised for growing crops in the greenhouse and collecting drainage water. The analyses of the latter indicated that an increase in fineness of division of pulverized limestone was responsible for a decreased loss of ammonia and nitrate nitrogen, thereby conserving the nitrogen supply. There was an increased loss of calcium, however, which is an important consideration from the standpoint of economy in the application of lime.

6. As in the preceding experiments an increase in fineness of division of pulverized limestone was responsible for a corresponding decrease in the lime-requirement of the soil. The titration of the acidity and alkalinity of the drainage waters indicated that an increase in fineness of division of pulverized limestone was accompanied by a decrease in acidity as measured by N/10 NaOH and an increase in alkalinity as measured by N/10 HCl.

SUMMARY

Considering as a whole the results set forth in this investigation, some interesting facts have come to light which appear to have a direct bearing upon the agricultural practice of liming. The influence of fineness of division of pulverized limestone, upon crop yield on various soils has been discussed, together with the effect upon the chemical and bacteriological factors of the soil. The more important points which have been established under the limitations of the experiment may be summarized as follows.

1. An increase in fineness of division of pulverized limestone from 20 to 40, 60 to 80, 100 to 200, to finer than 200-mesh is responsible for a proportional increase in the yield and total nitrogen content of crimson clover on several soil types (representing extensive fertile areas in the United States); and a corresponding decrease in lime-requirement.

2. From the above standpoint there was little choice between burnt lime and 200-mesh limestone.

3. Employing an apparatus devised for measuring the rate of neutralization of soil acidity by different grades of pulverized limestone, it was found that in three different soils the limestone finer than 60-mesh required about three weeks, while 20-mesh limestone required more than seven weeks to effect neutralization. An increase in the quantity of limestone required for neutralization was proportional to an increase in fineness of division of pulverized limestone, for any given period of time.

4. An increase in fineness of division of pulverized limestone is responsible for an increase in the activity of the bacteriological processes of ammonification, nitrification and nitrogen fixation (within certain limitations) as measured in soil and solution. This holds true regardless of whether the amount of limestone applied is less or somewhat more than the indicated Veitch lime-requirement.

5. The increase in bacteriological activities with an increase in fineness of division of pulverized limestone may be correlated directly with the chemical factors involved, i.e., the rate of neutralization of acidity, etc., and also crop yield.

6. With pots cropped to barley, buckwheat and rape (twice) which had been designed to permit the collection of drainage water, it was found that on a light open sandy loam the highest yields were obtained from soils treated with 60 to 80-mesh limestone. Two hundred-mesh proved superior to 20-mesh limestone but inferior to the former, because the material was probably washed down below the root zone. These results were paralleled both in the series with and without nitrogen, although the yields in the former case were superior to those in the latter. However, the fine limestone without nitrogen gave almost as high a yield in several instances as the coarse material with an application of 660 pounds of ammonium sulfate to the acre. The highest average per cent of nitrogen, however, was obtained with 200-mesh limestone.

7. An increase in the fineness of division of pulverized limestone was accompanied by a reduction in the lime-requirement. An analysis of the drainage waters indicated that there was a decreased loss of ammonia and nitrate nitrogen but an increased loss of calcium.

8. In general, an increase in the fineness of division of pulverized limestone is responsible for a proportional increase in crop yield, as well as for exerting a beneficial influence on the chemical factors in the soil. Furthermore, 200-mesh limestone may be regarded as effective as burnt lime.

In conclusion, it is a privilege to express appreciation to Dr. J. G. Lipman for his valuable advice so generously rendered in the planning and execution of this investigation. The writer is also indebted to Professor A. W. Blair for his assistance and interest throughout the work; and to his colleagues in the laboratory for many timely suggestions. Thanks are due Dr. H. E. Kiefer, Mr. E. C. Carhart and Mr. E. Meyer for material furnished by them.

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PLATE I

Galvanized iron pots permitting collection of drainage water, cropped to barley.

Treatment: 11, 12, Minerals + 12 grams $(\text{NH}_4)_2\text{SO}_4$; 13, 14, Minerals + 12 grams $(\text{NH}_4)_2\text{SO}_4$ + 20-40 mesh limestone; 15, 16, minerals + 12 grams $(\text{NH}_4)_2\text{SO}_4$ + 60-80 mesh limestone.



THE YIELD AND NITROGEN CONTENT OF SOYBEANS AS INFLUENCED BY LIME¹

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In earlier papers (2, 4) the authors have called attention to some of the factors which may influence the protein content of soybeans. It was pointed out, for example, that the shelled beans from limed plots contained a higher percentage of nitrogen than those grown on similar unlimed plots, and that this was true for all the varieties experimented with.

A continuation of the work has confirmed the earlier results, and has further shown that what is true of the shelled beans is also generally true of the top part of the plant when harvested as forage or as dry stalks, and of the roots with their accompanying nodules.

In this connection it was suggested by the senior author that a count of the nodules on the roots of plants from limed and unlimed plots might throw some light on the subject.

METHOD OF CONDUCTING THE EXPERIMENT

The soil on which this experiment was carried out is a loam of fair quality, but previous to 1908 it had been neglected. In the spring of 1908 certain of the plots received a treatment of ground limestone at the rate of 2000 pounds per acre, and in 1913 a further treatment at the rate of 4000 pounds per acre. Other plots referred to as the "unlimed plots" have received no lime treatment. All plots receive annual applications of acid phosphate at the rate of 400 to 600 pounds per acre and muriate of potash at the rate of 100 to 200 pounds per acre. Since 1908 the plots have been used for leguminous crops almost entirely and as this crop has been soybeans for the past four years the soil is now well inoculated, as shown both by luxuriant growth and the abundance of nodules on the roots. The soil originally contained about 0.09 to 0.10 per cent of total nitrogen. The beans were planted about May 8 in rows 33 inches apart. Germination was good and during the season they received the usual attention in the way of cultivation and hoeing.

RESULTS FROM CROP HARVESTED AS FORAGE

When the pods were well set, but before ripening had commenced, twelve plants were carefully removed from one of the rows of each of 6 varieties that

¹ The authors are indebted to Mr. H. C. McLean for the chemical analyses and to Mr. L. K. Wilkins for supervision of the field work.

were growing on limed and unlimed plots, and a count was made of the number of nodules per plant. This count showed an average of 83.6 nodules per plant for the six varieties grown on limed plots, and 50 nodules per plant for the same varieties on unlimed plots.

The number of nodules varied rather widely with the different varieties, but with all varieties the number was greater on the limed than on the unlimed plots. This would seem to substantiate further the generally accepted view that well limed soils, other things being equal, present more favorable conditions for inoculation, and therefore for the accumulation of atmospheric nitrogen by means of leguminous crops, than soils that are distinctly acid. The results obtained from the six varieties harvested as above described are given in table 1.

With one exception only, the yield of dry matter, both tops and roots, is greater for the limed than for the unlimed plots. Likewise, with only one exception, the percentage of nitrogen in the dry matter, both tops and roots, is greater for the limed than for the unlimed plots.

The average nitrogen content of the tops for the six varieties is—limed, 3.08 per cent, and unlimed 2.67 per cent; and the average for the roots is—limed, 1.47 per cent, and unlimed, 1.24 per cent. It is thus shown that decidedly more nitrogen has been accumulated on the limed than on the unlimed plots. This is shown in the two columns at the right—the average amount in a single plant, root and top, for the limed plots being 0.71 gram and for the unlimed plots 0.43 gram.

Manchu for the limed plots shows the highest percentage of nitrogen, both roots and tops, and also the largest amount of nitrogen recovered in the crop.

RESULTS FROM THE MATURED CROP

The beans on a portion of each one of the plots referred to above were allowed to ripen and were harvested by pulling up the entire plant. These were stood in bunches in the field until dry, when they were taken in and threshed and samples of both beans and stalks prepared for analysis. At the time of harvesting, the plants had shed practically all of the leaves so that these were left on the field in place of the roots. With the stalks were included the roots and also the bean hulls.

The dry weights, calculated to the acre basis, together with the results of the nitrogen determinations are shown in table 2.

Here, as with the forage, the yield of dry matter and percentage of nitrogen for both shelled beans and stalks, are, with slight exception, higher on the limed than on the unlimed plots. This, it may be pointed out, is in accordance with results secured in similar experiments conducted in 1914 (4) and 1915 (2).

The average yield of shelled beans for the six varieties (Cloud, Hollybrook, Ito San, Manchu, Ohio 9035 and Swan) on the limed plots is 19.3 bushels

TABLE I
The influence of time on the number of nodules and percentage of nitrogen for soybeans when harvested at half

TABLE 2
The influence of lime on the yield and nitrogen content of soybeans when harvested at maturity
Calculated on the acre basis

VARIETY	SOYBEAN SEEDS						SOYBEAN STEALS				TOTAL NITROGEN RECOVERED IN SEEDS AND STEALS	
	Unlimed			Limed			Unlimed		Limed		Unlimed	
	Dry matter bushels per acre pounds	Nitrogen bushels per acre pounds	Dry matter bushels per acre pounds	Dry matter bushels per acre pounds	Nitrogen bushels per acre pounds	Dry matter bushels per acre pounds	Dry matter pounds	Nitrogen pounds	Nitrogen pounds	Dry matter pounds	Nitrogen pounds	Nitrogen pounds
Baird.....	20.80	7.025	87.66	13.13	5.540	43.68	3,552	1.107	39.30	2,012	0.499	10.04
Cloud.....	19.00	5.744	65.94	15.13	6.023	54.72	2,052	0.608	12.48	1,492	0.479	126.96
Ebony.....	19.90	6.598	78.78	15.13	6.394	83.82	2,406	1.626	39.12	2,670	0.788	117.90
Edna.....	21.85	6.394	83.82	15.13	6.023	54.72	2,589	0.907	23.46	2,344	0.718	107.28
Guelph.....	18.40	5.698	62.88	15.13	6.412	54.88	2,492	0.618	15.52	2,670	0.788	78.30
Hollybrook.....	14.27	6.032	63.32	12.27	5.642	41.52	2,344	0.718	16.80	1,492	0.479	7.15
Ito San.....	17.50	6.032	63.32	12.27	5.642	41.52	2,670	0.788	21.04	1,492	0.479	71.68
Manchu.....	19.80	6.561	77.92	12.27	5.642	41.52	1,612	0.519	8.40	864	0.608	84.36
Manhattan.....	16.25	6.236	60.78	11.33	6.245	42.48	2,325	0.758	17.64	2,120	0.628	1,228
Medium yellow.....	18.00	5.837	63.04	12.87	5.522	42.64	2,344	0.718	13.28	1,536	0.947	14.55
Ohio 9035.....	23.06	5.884	81.44	14.40	5.392	46.56	2,216	1.446	32.08	1,536	0.947	11.52
Swan.....	21.67	6.461	84.00	11.33	6.245	42.48	1,900	0.828	15.76	920	0.539	9.76
Tarheel.....	9.83	5.642	33.28	11.33	6.245	42.48	4,610	0.967	44.56	2,667	0.618	77.84
Wilson.....	25.55	5.726	87.78	11.33	6.245	42.48	2,667	0.618	16.50	2,667	0.618	104.28
Average.....	19.30*	6.150*	71.20*	13.19	5.727	45.27	2,041*	0.791*	16.47*	1,342	0.615	8.26
												87.67*
												53.52

* Average of the six varieties having corresponding unlimed plots. The other varieties did not have corresponding unlimed plots.

per acre and for the same varieties on the unlimed plots it is 13.2 bushels per acre. The increase on the limed plots of 6.1 bushels per acre would more than pay for the lime, but since this is the fourth crop of beans since the last application of lime and all the crops were benefitted by the liming, the lime has been paid for long ago, and the 6.1 bushels represent net profit in addition to the increase in amount of stalks, which have some value. In 1915 seven varieties on limed plots yielded, on an average, 6.5 bushels more than the same varieties on unlimed plots.

Out of the fourteen varieties in 1916, only four fell below a yield of 18 bushels per acre on the limed plots, while the Wilson gave a yield of 25.5 bushels and Ohio 9035, Swan, Edna and Baird yielded between 20 and 25 bushels per acre. The low yield secured with Tarheel must no doubt be attributed to the fact that this is a slow-growing late-maturing variety; had heavy frosts come early the yield would have been less, and possibly no beans would have matured. This variety, however, if given a long growing season, makes a heavy stem and leaf growth and for this reason would make an excellent green-manure crop where land is not to be otherwise used during the summer and fall. The heavy growth is indicated by a yield of over two tons per acre of dry stalks. The yield of dry stalks is, without exception, larger on the limed than on the unlimed plots. For the six varieties that have corresponding unlimed plots, the yield is approximately 50 per cent greater, while the total nitrogen for these same varieties is just about 100 per cent greater.

With only one exception (stalks from Manchu), the percentage of nitrogen in beans and stalks from limed plots is greater than it is from corresponding unlimed plots. This is in accord with the results of the past three years, reference to which has already been made. This is likewise in accord with results secured at the Massachusetts Agricultural Experiment Station with red clover (5).

It is thus shown that the use of lime has consistently increased the yield of beans and stalks and has also given a feeding material richer in protein than that grown on unlimed plots. Special interest attaches to the total nitrogen recovered in seeds and stalks for the limed and unlimed plots. The lowest yield for a limed plot—71.68 pounds per acre—is greater by about 10 pounds than the highest yield on an unlimed plot.

The following varieties, limed, gave a yield of nitrogen above 100 pounds: Baird, Ebony, Edna, Ohio 9035 and Wilson, with Swan almost at the 100-pound mark. The average for the six unlimed plots is 53.52 pounds, while the average for the six corresponding limed plots is 87.67, or an increase of more than 50 per cent.

It is not possible to say how much of this nitrogen was gained from the air, and how much from the soil. It has been shown, however, from a number of experiments covering a period of eight or nine years (3) that non-leguminous crops grown on nearby plots, where the soil is practically the same as that of the soybean plots, when unaided by nitrogenous fertilizers or

green manures, are not able to gain from this soil more than about 20 to 25 pounds of nitrogen per acre, and while it does not necessarily follow as true, it seems reasonable to assume that the soybeans would probably not take from the *soil* more nitrogen than the other crops. This would mean, on an average, about 65 pounds of nitrogen per acre taken from the air, and if allowance is made for the leaves which were left on the field, it would probably mean more. This is in fair agreement with results secured by means of cylinder experiments in an indirect way, where leguminous crops were used as a source of nitrogen in comparison with nitrate of soda and stable manure (1).

This points to the soybean as a plant especially efficient as an accumulator of atmospheric nitrogen and emphasizes the desirability of its wider use as a means of maintaining the nitrogen and humus supply of the soil.

SUMMARY

1. Earlier experiments have shown that when soybeans are grown on limed and unlimed plots, the liming not only increases the yield of dry shelled beans, but also the nitrogen content of the beans.
2. The results secured in 1916 confirm the earlier experiments and also show that liming increases the yield of forage when the crop is harvested either as hay or as dry stalks, and also increases the percentage of nitrogen in these materials.
3. A count of the nodules on roots of plants taken up about the time the pods were fairly well filled showed an average of 83.6 nodules per plant on the limed plots and 50 nodules per plant on the unlimed plots.
4. The average yield of shelled beans on the unlimed plots was 13.2 bushels per acre and the average on the corresponding limed plots was 19.3 bushels. The average percentage of nitrogen in the beans for unlimed plots was 5.73 per cent and for the limed plots 6.15 per cent. In 1915 the corresponding percentages were 5.92 and 6.41, and in 1914 they were 6.21 and 6.70.
5. The average yield of stalks was, for unlimed plots, 1342 pounds per acre, and for corresponding limed plots, 2041 pounds per acre. The average nitrogen content of stalks from the unlimed plots was 0.615 per cent and from the corresponding limed plots, 0.791 per cent.
6. The average total yield of nitrogen recovered in the crop from the unlimed plots was 53.52 pounds per acre and the average for the corresponding limed plots 87.67 pounds per acre.
7. Reasoning from the amount of nitrogen which non-leguminous crops, without the aid of commercial fertilizers or green manures, have drawn from nearby plots where the soil is similar, it is believed that as much as 65 pounds of this nitrogen was taken from the air.

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FERMENTATION OF MANURE TREATED WITH SULFUR AND SULFATES: CHANGES IN NITROGEN AND PHOSPHORUS CONTENT

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INTRODUCTION

The loss of nitrogen from decomposing manure under certain conditions of fermentation proceeds rapidly. Wagner (21) has shown that the nitrogen of urine can be transformed into ammonium compounds within 48 hours after being voided. Pfeiffer (14), Wildt (17) and Deherain (7) have reported results showing that under certain conditions of fermentation manures may lose as much as 42 per cent of their total nitrogen as the free element. Niklewski (13) states that doubt is unfounded as to the liberation of elementary nitrogen from decomposing manure in consequence of the co-operation between nitrifying and denitrifying bacteria. Dietzell (8), Bjorn-Anderson (2) and Pfeiffer (14) have proved that the addition of acid phosphate to manure will prevent the liberation of nitrogen. Gerlach (9) has recently reported that the action of gaseous ammonia upon acid phosphate results in the formation of ammonium sulfate and calcium phosphate. This is evidence that the conservation of nitrogen in manure treated with acid phosphate is the result of a combination between ammonia formed and calcium sulfate furnished by the acid phosphate, rather than the prevention of the formation of ammonia.

The treatment of manure with gypsum is also a common practice. Experiments in which this material was added to both solid and liquid manure have been reported by Schneidewind (15), Severin (16), Dietzell (8) and Bohme (3). The results show that a loss of more than 50 per cent of the total nitrogen may occur from untreated manure, but that when 5 to 10 per cent of calcium sulfate is added a very small loss of nitrogen occurs and that the nitrogen, in the case of urine, is almost completely changed to ammonium sulfate, and held as such. Work reported by the Copenhagen Station (6) and by Stoklasa (18) shows that the addition of sulfuric acid reduced the loss of nitrogen from 75 per cent in untreated manure to almost nothing. Hall (10) and Schneidewind have each called attention to the harmful effects that may be caused by the application of gypsum-treated manure to the soil. The claim is made that sulfides produced by reduction of sulfate are poisonous

to growing plants. The results from the barnyard manure test plots of the Ohio Agricultural Experiment Station, where gypsum mixed with manure at the rate of 40 pounds per ton has been used since 1897, show no such effect, but, on the other hand, a gain over the plots treated with manure alone. It is also shown by these results (reported in Circular 144, Ohio Agr. Exp. Sta.) that the manure designated as "stall manure" has produced larger crop yields than "yard manure," thus proving that prevention of loss by leaching is necessary to realize the fullest return from reinforcing materials.

The use of elementary sulfur as a reinforcing material for manure has not as yet received much attention. Street (19) in 1900 reported experiments planned for the purpose of studying changes which nitrogen of barnyard manure undergoes in presence of various fertilizing materials. Results of this work continued during two years showed that sulfur either in the form of commercial flowers of sulfur or as sulfate of iron has a marked effect in decreasing the loss of ammonia.

Cook and Hutchinson (5) report that the addition of 1 pound of sulfur per bushel of manure greatly reduced the number of flies hatched from manure, but that such heavy applications applied to the soil would undoubtedly have an injurious effect on plant growth.

J. C. Lipman and associates (11) have recently called attention to the benefit to be gained from treatment of compost heaps with sulfur and "floats" by furnishing available phosphorus.

EXPERIMENTAL

For the purpose of studying the influence of sulfur and other materials upon the fertilizing value of manure as measured by changes in its composition and the response made by growing crops, experiments with reinforcing materials were started at the Ohio Agricultural Experiment Station in May, 1915. The first experiment reported deals with the effects of sulfur, calcium sulfate and acid phosphate upon the changes occurring in solid horse manure. Data pertaining to nitrogen, phosphorus and sulfur are included. Manure which is being treated in larger containers and under the conditions described is being applied after fermentation to small plots to determine the influence of the several treatments on soil conditions and plant growth.

Under the second experiment the effect of the same reinforcing materials on the nitrogen content of cow's urine is shown.

Experiment with solid manure

The horse manure used in this experiment had accumulated for about one week in box stalls, fine cut wheat straw being used as litter to facilitate mixing and sampling. The manure was mixed and all lumps pulverized to insure a complete intermixture of added materials. Four lots of about 30 pounds each were weighed out and treated as follows: to no. 1 was added

342 grams of acid phosphate; to no. 2, 407 grams of calcium sulfate; and to no. 3, 90 grams of flowers of sulfur; the fourth lot was left untreated. After addition of the different materials the samples were again thoroughly mixed and appeared to be of uniform structure. About 25 pounds of each of these mixtures was placed in weighed containers and the remainder saved for analysis.

The containers were 15 inches in diameter and 19 inches high, and were made with a cover that allowed a paraffin seal to be made. The only air admitted was through a glass tube extending to the bottom of the container through a rubber stopper in the center of the cover. The weight of container and contents was then obtained. Air was drawn through the apparatus for

TABLE 1
Changes in weight and loss of dry matter

	MANURE AND ACID PHOSPHATE	MANURE AND SULFUR	MANURE AND CALCIUM SULFATE	MANURE UNTREATED
	GRAMS	GRAMS	GRAMS	GRAMS
Total weight of mixture at beginning.....	8,471	11,632	11,726	11,211
Total weight of mixture at end.....	8,201	11,392	11,176	10,505
Total weight of dry matter at beginning.....	3,617	4,210	4,362	4,047
Total weight of dry matter at end.....	2,829	3,441	3,408	2,731
Percentage of dry matter lost.....	21.78	18.2	21.87	32.5
Total weight of moisture at beginning.....	4,854	7,422	7,364	7,164
Total weight of moisture at end.....	5,372	7,951	7,768	7,774
Gain in moisture.....	518	529	404	610
Percentage of moisture at beginning.....	57.3	63.9	52.8	63.9
Percentage of moisture at end.....	65.5	69.8	69.5	74.0

3 hours on 3 days a week by means of a Crowell rotary pump, which drew 100 liters per hour through each container. The air drawn through the containers was first passed through three wash bottles containing sulfuric acid and sodium hydroxide and water, in the order named, and then conducted through the glass tubes to the bottom of the containers. The air was drawn from the outlet in the top of the container and passed first into a trap and then through sulfuric acid and sodium hydroxide solutions for absorption of ammonia and hydrogen sulfide.

Loss of organic matter. After standing at room temperature from May 7, 1915 to January 14, 1916, the cans and contents were weighed and moisture determinations made immediately. The data for moisture content and loss of organic matter are given in table 1.

The results show that there was an appreciable loss of dry matter due to the destructive processes occurring during the fermentation of manure under the conditions to which it was subjected in this experiment.

The manure treated with flowers of sulfur lost only 18.2 per cent of its dry matter as compared with a loss of 32.5 per cent from the untreated manure. The samples treated with acid phosphate and calcium sulfate lost approximately the same amounts and only slightly more than the sulfur-treated manure. This is a reduction of more than 44 per cent in the loss of dry matter from the manure when treated with sulfur, and a reduction of more than 32 per cent by the addition of either acid phosphate or calcium sulfate.

These treatments appear to have been unfavorable for the development of cellulose-dissolving bacteria, which rapidly destroy cellulose (12), the products formed by their activities being immediately used by other microorganisms with the liberation of carbon dioxide and methane.

TABLE 2
Nitrogen loss and changes

	MANURE AND ACID PHOSPHATE	MANURE AND SULFUR	MANURE AND CALCIUM SULFATE	MANURE UNTREATED
	grams	grams	grams	grams
Weight of total nitrogen at beginning.....	66.0738	94.8008	89.1176	96.4146
Weight of total nitrogen at end.....	63.9678	91.1360	86.0552	86.1410
Weight of nitrogen lost.....	2.1060	3.6648	3.0624	10.2321
Weight of nitrogen recovered as ammonia.....	0.0026	0.0103	0.0296	0.0415
Weight of nitrogen lost as free element or ex- perimental error.....	2.1034	3.6545	3.0328	10.2321
Percentage of total lost.....	3.1	3.8	3.4	10.6
Weight of water-soluble nitrogen at beginning	30.83	42.80	43.85	41.14
Weight of water-soluble nitrogen at end.....	6.31	14.69	14.52	12.81
Percentage of original water-soluble nitrogen changed.....	79.53	65.6	66.88	68.8

A dried and ground sample of the sulfur-treated manure at the end of the experiment gave an acid water extract, while that of the untreated manure gave an alkaline reaction. The fact that the manures contained more water at the end of the experiment than at the beginning indicates that the bacteria had optimum working conditions so far as the moisture content was concerned.

Changes in nitrogen content. The amounts of different forms of nitrogen found in the manure before and after fermentation are given in table 2. Since a small amount only of the total nitrogen content was evolved as ammonia from the untreated and treated manure, it appears that the conditions of fermentation existing during the experiment were not favorable for the liberation of appreciable quantities of ammoniacal nitrogen. The weights of nitrogen evolved from the different samples as ammonia and as free nitrogen are shown in table 2.

Total nitrogen. The three lots of treated manure have lost approximately the same amounts of total nitrogen, viz., 3.5 per cent. This figure compared with a loss of 10.5 per cent in the untreated manure shows that there was a conservation of nitrogen by the reinforcing materials used. Since only a small amount of ammonia was recovered, nitrogen must have been lost as free nitrogen.

The treatments either have made conditions unfavorable for ammoniacal fermentation or have been efficient in preventing the escape of ammonia which may have been produced. The latter view appears to furnish the more correct explanation when the results obtained are considered in more detail.

The acid phosphate treatment, no doubt, furnished sufficient calcium sulfate to combine with ammonia as it was produced, forming ammonium sulfate. A reaction may also have taken place to some extent between monocalcium phosphate and ammonia. In the sample treated with calcium sulfate the ammonia formed would also be held as ammonium sulfate. Evidence of the effectiveness of calcium sulfate in this respect is furnished by results obtained in the second experiment reported where liquid manure was treated with calcium sulfate. The fact that the manure treated with sulfur developed acidity equivalent to 24 grams of sulfuric acid during fermentation is evidence that this acidity may have been responsible for the retention of nitrogen as ammonia where sulfur was used in this experiment. The conditions maintained were evidently such that the loss of free nitrogen was brought about by denitrification processes developing between periods of aeration or during the latter part of the experiment, during which time the containers were aerated to a less extent than previously.

Water-soluble and non-proteid nitrogen. There is, under all conditions of manure fermentation, a transformation of water-soluble nitrogen to insoluble nitrogen, which is due to the utilization of ammonia and other soluble compounds of nitrogen by bacteria. In this experiment the effect of bacterial activities during fermentation of the untreated manure and that treated with gypsum and sulfur has been to change approximately 67 per cent of the water-soluble nitrogen originally present to some form insoluble in water. In the acid phosphate-treated manure there was a greater decrease in the water-soluble nitrogen content, about 80 per cent being changed to insoluble nitrogen. The supply of soluble phosphorus furnished may have favored the development of bacteria.

During fermentation about 70 per cent of the non-proteid nitrogen present in the untreated manure and that treated with sulfur and calcium sulfate at the beginning of the experiment has been changed to proteid nitrogen.

The decreased content of water-soluble nitrogen and the increase in proteid nitrogen shows the effect of fermentation on solubility and availability of the nitrogen compounds of manure.

The work of R. A. Berry (1) regarding these changes in nitrogen content

of manure during storage shows that the solubility of nitrogen is decreased considerably during fermentation.

Changes in phosphorus. The analytical data pertaining to changes in the solubility of phosphorus in the different lots of manure are given in table 3. With the exception of the manure treated with acid phosphate, the total phosphorus content is approximately the same.

Water-soluble and citrate-insoluble phosphorus. During the fermentation of untreated manure the solubility of the phosphorus was decreased 24 per

TABLE 3
Changes in phosphorus

	MANURE AND ACID PHOSPHATE	MANURE AND SULFUR	MANURE AND CALCIUM SULFATE	MANURE UNTREATED
	grams	grams	grams	grams
Weight of phosphorus before and after fermentation.....	27.72	14.55	14.28	14.12
Weight of water-soluble phosphorus at beginning.....	10.81	6.53	5.12	5.68
Weight of water-soluble phosphorus at end.....	4.05	4.23	2.21	4.28
Percentage of original changed.....	62.53	35.2	56.83	24.6
Weight of phosphorus soluble in 0.2 per cent alkali at beginning.....	6.59	9.05	10.00	8.57
Weight of phosphorus soluble in 0.2 per cent alkali at end.....	2.46	5.32	3.09	5.09
Percentage of original changed.....	62.67	41.2	69.10	40.6
Weight of phosphorus soluble in 0.2 per cent acid at beginning.....	21.92	4.57	4.24	3.74
Weight of phosphorus soluble in 0.2 per cent acid at end.....	29.79	7.69	7.77	8.69
Weight of inorganic phosphorus at beginning.....	8.18	2.12	2.35	1.94
Weight of inorganic phosphorus at end.....	12.23	6.47	2.40	5.03
Weight of citrate-insoluble phosphorus at beginning.....	4.38	2.88	2.84	3.54
Weight of citrate-insoluble phosphorus at end.....	3.77	2.78	2.30	2.13
Percentage of original changed.....	13.92	3.4	19.01	40.0

cent. The reduction of water-soluble phosphorus in the manure treated with acid phosphate was 62 per cent of the soluble phosphorus present at the beginning of the experiment. As the acid phosphate added contained 5.8 per cent of water-soluble phosphorus, the greater decrease in solubility was that of soluble phosphorus added. Calcium sulfate has actively decreased the solubility of phosphorus originally present. If depression of solubility of phosphorus in fermenting manure is due to the function which phosphorus performs in the life cycle of bacteria, it would seem that calcium sulfate had stimulated bacterial activities.

The amount of citrate-insoluble phosphorus in all the samples was less after fermentation than before, showing that during fermentation there has been a change from insoluble to citrate-soluble forms. The greatest change is found in the untreated sample, in which 40 per cent of the original citrate-insoluble phosphorus has been changed to forms soluble in neutral ammonium citrate solution. The acid phosphate and calcium sulfate samples show a change of 13 and 19 per cent, respectively; the change in manure treated with sulfur was very small. The results for water-soluble and citrate-insoluble phosphorus show that, while there has been a loss of water-soluble phosphorus during fermentation, at the same time there has occurred an increase of phosphorus soluble in neutral ammonium citrate. Similar results are reported by Tottingham and Hoffman (20).

Inorganic phosphorus. The results obtained for inorganic phosphorus by the method (4) used show that the amount present at the beginning of the experiment has been increased during fermentation, except in the case of the calcium sulfate-treated manure, where the same amount was found at the end of the experiment as at the beginning.

Solubility of phosphorus in 0.2 per cent sodium hydroxide solution has been reduced in all the samples by approximately 50 per cent of the amount originally present. Aside from having a solvent action on iron and aluminum phosphates possibly present, it would be expected that the action of dilute alkali would be greater on organic phosphorus compounds than on inorganic phosphates. There has been a decidedly increased solubility of phosphorus in 0.2 per cent hydrochloric acid, and this increase has been greater in the manure to which no phosphorus was added.

The comparative solubility of phosphorus in 0.2 per cent hydrochloric acid and water, before and after fermentation, is of interest as indicating a change from organic to inorganic phosphorus. With the exception of manure treated with acid phosphate, less phosphorus was soluble in 0.2 per cent acid than in water at the beginning of the experiment, while after fermentation the acid-soluble phosphorus exceeded the water-soluble. The reduced solubility of phosphorus in dilute alkali and the increased solubility in acid, considered in connection with results obtained for inorganic phosphorus, furnish evidence of the change from organic to inorganic phosphorus during the period of fermentation, under the conditions of this experiment.

Changes in sulfur. Considerable quantities of sulfur were evolved as hydrogen sulfide from the treated and untreated manure during fermentation. The formation of hydrogen sulfide, however, proceeded at a much slower rate in the untreated than in the manures treated with sulfur, calcium sulfate and acid phosphate. A measure of the hydrogen sulfide evolved was obtained by determining the sulfur content of the sodium hydroxide solutions used in the absorption of hydrogen sulfide. The weights of sulfur which were evolved as hydrogen sulfide were as follows: From manure treated with sulfur, 9.29 grams; manure treated with acid phosphate, 6.01 grams; manure

and calcium sulfate, 3.46 grams; and from the untreated manure, 0.3867 grams. These figures show that the largest amount for hydrogen sulfide was obtained from the sulfur-treated manure.

The odor of the air exhausted from the containers indicated that organic compounds were being evolved and not absorbed by the acid and alkali solutions through which the air was passed.

Determination of water-soluble sulfates in the sulfur-treated manure showed the presence of sulfates equivalent to 15.2 grams of sulfuric acid at the beginning and 38.6 grams at the end of the experiment. This is a production of sulfates equivalent to 23.4 grams of sulfuric acid during the period of fermentation. The untreated manure showed a loss of sulfates equivalent to 4 grams of sulfuric acid which is due in part to the evolution of hydrogen sulfide.

The results obtained by titrating the boiled water extracts of the sulfur-treated and untreated manure with tenth-normal alkali solution gave the same indications as were obtained by the determination of water-soluble sulfates in two samples. Water extracts of both the sulfur-treated and untreated were acid in reaction at the beginning of the experiment, while after fermentation the sulfur-treated manure had increased in acidity and the untreated manure had become alkaline in reaction.

Experiment with liquid manure

The effect of sulfur, calcium sulfate and acid phosphate on liquid manure was studied under the following conditions: 500-cc. portions of cow's urine were placed in 2-quart jars and the materials referred to added in amounts as shown in table 4. The jars were covered with cheesecloth and placed in

TABLE 4
Changes in nitrogen content of liquid manure

TREATMENT	WEIGHT OF NITROGEN AT BEGINNING	WEIGHT OF NITROGEN AT END	LOSS	WEIGHT OF AMMONI- ACAL NITROGEN AT BEGINNING	WEIGHT OF AMMONI- ACAL NITROGEN AT END	GAIN
	grams	grams	per cent	grams	grams	grams
27 grams acid phosphate.....	5.09	4.83	5.1	0.2	0.4642	0.2642
7 grams sulfur.....	5.09	4.58	10.0	0.2	0.2923	0.0923
27 grams calcium sulfate.....	5.09	4.595	9.72	0.2	3.4750	3.2750
Untreated.....	5.09	0.989	80.58	0.2	0.4299	0.2299

a room free from ammonia fumes and allowed to stand from July 8 to August 14, 1916. The contents of the jars were stirred at regular intervals during this period. The results reported showing a loss of total nitrogen and the changes in ammoniacal nitrogen occurring in the several portions are the average of duplicate determinations upon duplicate jars.

The untreated urine lost 80 per cent of its total nitrogen. While the additions of sulfur, calcium sulfate and acid phosphate have considerably decreased this loss, the results show that acid phosphate has been most effective in this respect.

The quantities of ammoniacal nitrogen found in the treated portions of urine, as compared with the ammoniacal nitrogen content of the untreated portion at the end of the experiment, afford an explanation of how the treatments have operated in preventing loss of nitrogen. In the case of urine treated with calcium sulfate, the weight of ammoniacal nitrogen at the end of the experiment was only slightly less than the total nitrogen content, indicating that ammonia was transformed into ammonium sulfate and held in this form as rapidly as it was produced.

As the ammoniacal nitrogen content of acid phosphate-treated urine at the end of the experiment is only slightly increased over that of the untreated portion, acid phosphate has prevented the formation of an appreciable amount of ammonia during the period from July 8 to August 14. The same conclusion can be drawn from the results for the sulfur-treated urine.

Some time after the period during which the experiment was continued, the formation of ammonia in the acid phosphate-treated portion developed rapidly. After the removal of a part of the contents of the jars on August 14 for determination of total and ammoniacal nitrogen the jars were sealed and allowed to stand until December 7. Upon opening the jars at this time the acid phosphate-treated sample was found to be alkaline and evolving ammonia at such a rapid rate that red litmus paper suspended in the jar was quickly changed from red to blue. This is of considerable interest in view of the fact that acid phosphate was most effective in decreasing loss of total nitrogen, and at the same time prevented the formation of ammoniacal nitrogen during the period from July 8 to August 14.

SUMMARY

The loss of dry matter from manure after fermenting for 250 days was 32.5 per cent in untreated manure and 21.8 per cent from manures treated with acid phosphate and calcium sulfate, while the sulfur-treated manure lost 18 per cent.

Manures treated with acid phosphate, sulfur and calcium sulfate lost approximately 3.5 per cent of their total nitrogen as compared with a loss of 10.5 per cent from the untreated manure.

The water-soluble and non-protein nitrogen were greatly reduced during fermentation.

The water-soluble phosphorus has been decreased in all the samples, but at the same time the citrate-insoluble has also been decreased.

The solubility of phosphorus in 0.2 hydrochloric acid has been increased, and the organic phosphorus was greatly decreased during fermentation.

The three treated manures evolved large amounts of hydrogen sulfide. The largest amount was evolved from manure to which sulfur was added.

The manure treated with flowers of sulfur produced water-soluble sulfates equivalent to 23.4 grams of sulfuric acid, as compared with a loss of about 4 grams from the untreated sample during fermentation.

The acidity of water extracts of untreated and sulfur-treated manure was the same at the beginning of the experiment, but during fermentation the sulfur-treated manure increased in acidity, while the untreated sample became alkaline.

Sulfur, calcium sulfate and acid phosphate were very effective in preventing loss of nitrogen from urine. The untreated urine lost 80 per cent of its total nitrogen.

Treatment with sulfur reduced the loss of nitrogen to 10 per cent and prevented formation of ammonium salts.

The calcium sulfate-treated sample lost 9.7 per cent of its nitrogen, and 68 per cent of its total nitrogen was transformed to ammonium sulfate and held as such.

The urine treated with acid phosphate lost only 5 per cent of its nitrogen, and the treatment prevented the formation of ammoniacal nitrogen in an open container during the 37-day period of the experiment.

After standing in a closed jar three months longer, the acid phosphate-treated urine was found to be alkaline and evolving ammonia.

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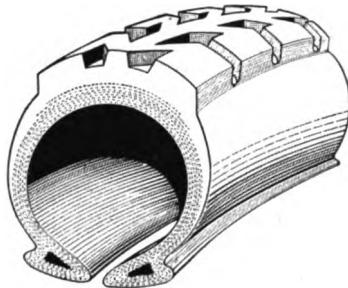
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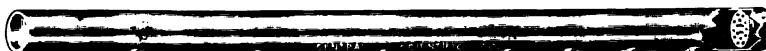
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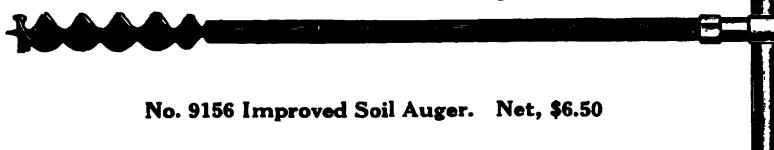
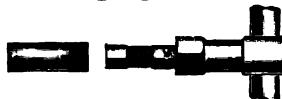
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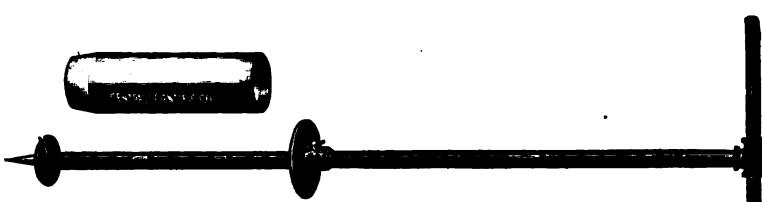
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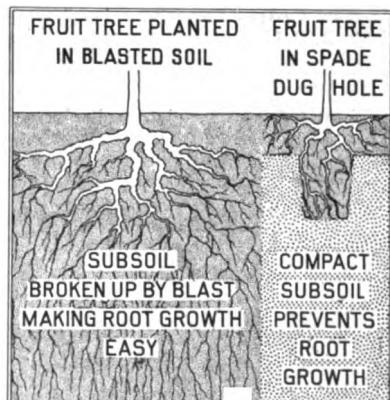
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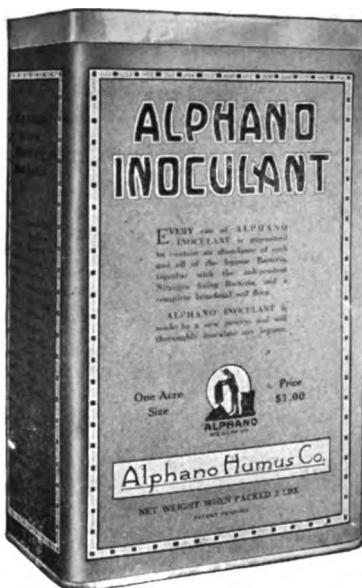
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A PRELIMINARY SOIL CENSUS OF ALABAMA AND WEST FLORIDA

ROLAND M. HARPER

Received for publication February 28, 1917

The state of Alabama is readily divisible into fifteen or twenty geographical divisions, differing in geology, soil, topography, vegetation, and cultural features. The soils of each of these were described, as accurately as was possible at that time, by Dr. Eugene A. Smith, state geologist, in the sixth volume of the Tenth Census, 1884, and in his report on the agricultural features of Alabama, published by the State at about the same time. These reports include chemical analyses of soils from forty-nine localities, but these are too few to be thoroughly representative of so many regions, and indeed some of the regions are not represented at all.

Comparatively little has been added to our knowledge of the chemistry of Alabama soils since 1884; but since 1902 the United States Bureau of Soils has made more soil surveys in Alabama than in any other state—no doubt largely on account of the splendid foundation laid by Dr. Smith—and over half the area of the state, including the whole of some geographical divisions and parts of all the others, has now been mapped by that organization.

West Florida, which is that part of Florida west of the Apalachicola River, is here combined with Alabama to make an area of more regular shape and to illustrate some geographical principles better.¹ Four of the divisions of Alabama extend into it, and it also has four smaller divisions which are not represented in Alabama. Three government soil surveys cover parts of it, and the soils of the whole state were described by Dr. Smith in the same Tenth Census volume above mentioned.

The present census is based on all the government soil surveys for Alabama and West Florida published up to the end of 1916. As is doubtless well known to readers of this magazine, these reports, most of which cover a single county, say little or nothing about soil chemistry, but classify soils according to origin, color, moisture, texture, etc. Except for a few essentially non-agricultural types, such as rock outcrop, swamp, and coastal beach, each type of soil is given a generic or series name, usually of geographical origin, denoting its supposed history and color, and a specific or class name indicating its texture.

¹ Ever since Florida became a part of the United States there has been more or less talk of annexing West Florida to Alabama; and the Methodists long ago included it in their Alabama Conference. Sections 81 and 82 of Bulletin W of the United States Weather Bureau together cover exactly the same territory as this paper.

As the work has progressed during the last fifteen years or so, the number of series names has multiplied enormously, until now the distinctions between some of them are pretty hard to grasp. This makes it impracticable to use the soil series for statistical purposes, for surveys made of similar or neighboring areas several years apart may differ greatly in the number of series recognized. Or different men working in similar areas at the same time might easily interpret the same type of soil differently.

Although the nomenclature of the older surveys is revised from time to time, and long lists of changes can be found in some of the publications of the Bureau of Soils, particularly Bulletin 96 (the last of its series), published about the middle of 1913, there seems to be no special incentive for keeping the revision always up to date, and indeed in many cases it cannot be done properly without re-examining the areas at considerable expense and subdividing some of the types as originally mapped into two or more.

Unlike the similarly named geological formations, the soil series names have not come into general use among scientists, so that they would be meaningless to persons not familiar with the publications of the Bureau of Soils; but possibly this will be changed with the lapse of time. Soil series are also unlike geological formations in not having a fixed position in a linear series, which is another obstacle to averaging them up in statistics.

When texture classes alone are considered, regardless of series, most of the difficulties just mentioned are avoided. The texture classes can be arranged pretty well in a linear sequence, and their names (e.g., loamy fine sand, silty clay loam) are self-explanatory. The number of such classes is not likely to increase much, and although there will always be some uncertainties of identification, owing to the complete intergradation between different types, the errors will tend to balance when several surveys are taken together.

Each soil survey of a county or similar area lists the various soil types in order of area and gives the approximate acreage of each, as well as indicating their distribution on the map, on a scale of an inch to the mile. A person familiar with the geographical boundaries, therefore, can apportion the different soil types among the respective regions and compute the percentage of each with reasonable accuracy. (This has already been done in a general way for the whole United States and its larger divisions on pages 9-10 of Bulletin 96 of the Bureau of Soils, and in more detail for the state of Virginia in Bulletin 46 of the United States Department of Agriculture published a few months later.)

In these two government publications each of the larger divisions, like the Piedmont region and the coastal plain, is assigned a different set of series names, so that one can pick out the soils of each division pretty well without looking at the maps; but in this paper the divisions used are considerably smaller, and where a county includes parts of two or more of the divisions careful scrutiny of the map is required. When the areas of all the soils mapped in a given region are determined (it took about three days of steady

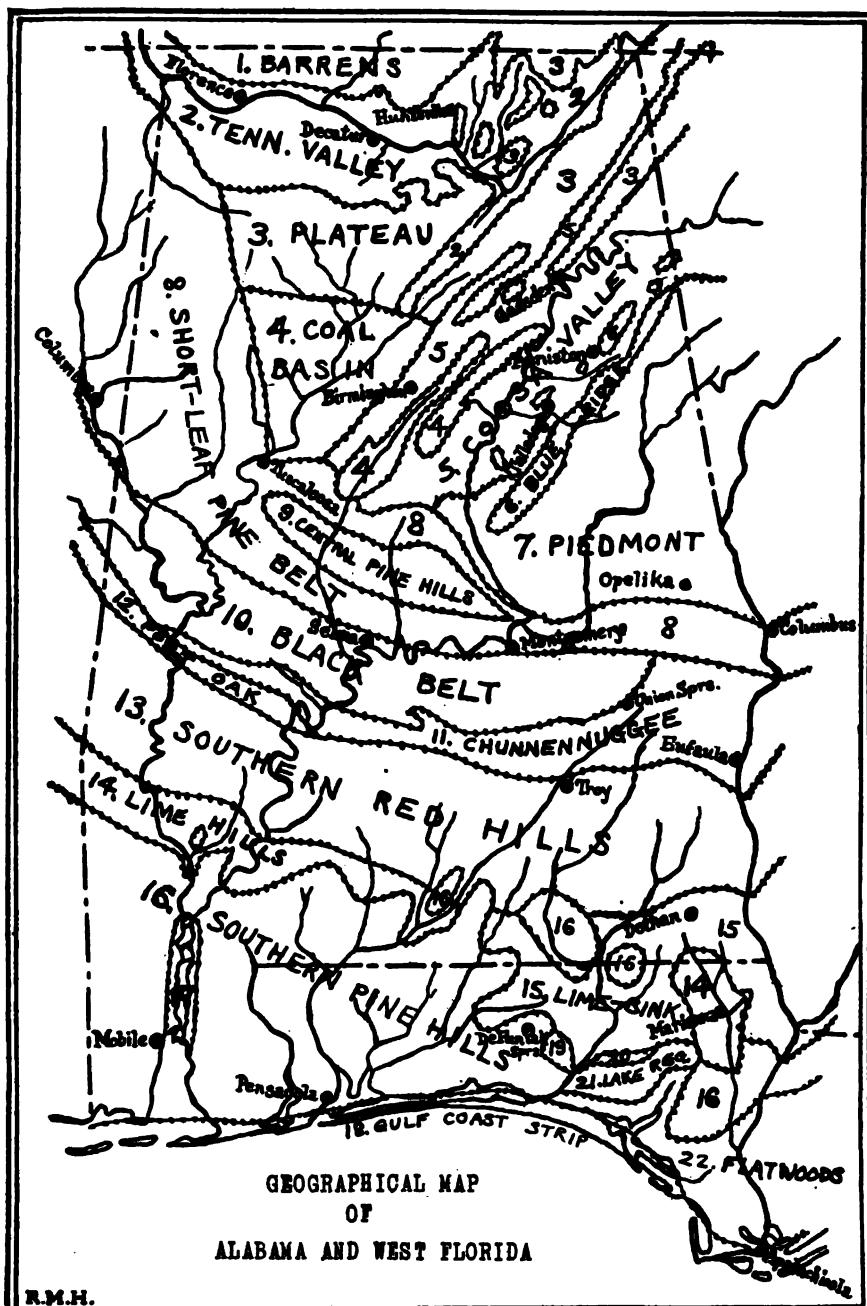


FIG. 1. MAP SHOWING THE REGIONS DESCRIBED HEREIN, AND ALSO THE PRINCIPAL RIVERS AND CITIES, TO ILLUSTRATE THEIR RELATIONS TO SOME OF THE GEOGRAPHICAL BOUNDARIES

Regions 17, 19 and 20, whose names are omitted from the map for lack of space, are the Mobile delta, the Knox Hill country, and Holmes Valley.

work to cover Alabama) they can be grouped together by texture classes, or by series if desired, and the percentages calculated.

The geography of Alabama and the northern part of Florida has been described by the writer in reports on forests published by the geological surveys of those states in 1913 and 1914, respectively, and the map used here-with shows very nearly the same geographical boundaries that were defined in those reports.² Some of these boundaries are so sharp that the transition from one region to the other is complete in less than a mile, but they have not all been located at a sufficient number of points to be mapped with perfect confidence. In some cases the boundary, though sharp enough, is too intricate to be drawn accurately on a map of this scale, and must therefore be generalized, as a topographic contour would be under the same circumstances. In other cases the transition from one region to another is so gradual that different persons might differ by a few miles in locating the boundary. On the present map no distinction is made between the well-defined and the indefinite boundaries, but some information on these points may be gathered from the geographical reports just mentioned, and from some of the soil surveys. One of the least satisfactory boundaries at the present time is that between the lime-sink region and the southern pine hills, principally because of lack of opportunity to trace out all its sinuosities. But fortunately this uncertainty does not materially affect the soil statistics, for these two regions do not differ as much in soil as they do in topography. Two divisions of the central pine belt of Alabama that were kept separate in 1913 are here combined, because their differences are mainly geological, and it is almost impossible to separate them by their soils alone.

Brief sketches of the geographical divisions now follow, beginning with the northernmost. The first seven belong to the highlands, or mineral region, and the rest to the coastal plain. The descriptions are on the following plan: area in Alabama and general distribution elsewhere, geology, topography, principal series and classes of soils in order of area, percentage of evergreens in the forests, commonest native trees in order of abundance, percentage of improved land in 1880, and the expenditure for fertilizers in 1909 per acre of improved land in 1910. The relative extent of the different soil series is not considered very important, for reasons previously mentioned, but may be of interest to persons familiar with the government classification, and to others who can easily remember some of the more important series, such as the reddish clayey "Orangeburg," the yellowish sandy "Norfolk," and the black mucky "Portsmouth."

The statistics of vegetation and agriculture are given as a contribution toward correlating such features with soil texture. Those who have investigated soils from a chemical standpoint have long known that deciduous trees

² A similar but less detailed map of Alabama and some of the same agricultural statistics were used by the writer in the Proceedings of the Society of American Foresters, vol. 11, p. 208-214. 1916.

in general and certain species in particular characterize the richer soils, and also that the more fertile the soil the larger proportion of it is cultivated and the less the expenditure for fertilizers.³ And regardless of chemical composition, the soils of finest texture are generally the most productive, except that the clays and mucks are often inferior to the loams and silt loams, on account of imperfect aeration.⁴ The vegetation statistics are taken with slight modification from the geographical reports cited. The statistics of improved land are computed from the 1880 census because that was more accurate than any earlier one, and because after that differences in original fertility were being overcome more and more by the use of fertilizers. The statistics of fertilizer consumption have been computed from the 1910 census, because that is the latest.

In these regional sketches, and also in the table following them, some soil classes which do not differ much are lumped together for the sake of simplicity, e.g., rock outcrop and rough stony land, stony, gravelly and cherty loam, shale and slate loam, and swamp, muck, and meadow.⁵

THE REGIONS

1. The "barrens" of the Tennessee valley, a continuation of the Highland Rim of Middle Tennessee, cover about 800 square miles in Alabama. The geological formation is mostly Keokuk chert, of Mississippian age, with strata horizontal or nearly so. The soils are mainly grayish or yellowish. The topography is comparatively level except in the vicinity of streams, which have cut valleys proportional to their size. The whole area in Alabama is covered by three soil surveys, in which the soils are referred mainly to the Clarksville series, with small amounts of Decatur, Hagerstown, etc. Silt loam, stony and gravelly loam, and clay loam cover about 99 per cent of the area. Silt loam is relatively more abundant here than in any other part of the state.

About 30 per cent of the forest is evergreen. The commonest trees are short-leaf (loblolly) pine, red oak, sweet gum, short-leaf pine, post oak, willow oak, and beech. About 25 per cent of the area seems to be cleared at present, and the annual expenditure for fertilizers probably does not exceed 50 cents an acre, though no accurate statistics can be given for such a small area, which does not cover the whole of any county.

³ See Harper, R. M. Vegetation and soils. *In* Science, n. s., vol. 42, no. 1084, p. 500-503. October 8, 1915.

⁴ In this connection see Cannon, W. A., and Free, E. E. The ecological significance of soil aeration. *In* Science, n. s., vol. 45, no. 1156, p. 178-180. February 23, 1917.

⁵ In the Bureau of Soils publications from the beginning the term "meadow" has been employed regardless of ordinary usage to designate almost any land subject to overflow from small streams, whether it bears meadow vegetation or not (in the South it is usually wooded). But many soils which in the earlier reports were called simply Swamp or Meadow are now assigned to definite series and texture classes, so that the percentage of swamp land as determined from the soil surveys is only a rough approximation.

2. The Tennessee valley proper lies on both sides of the Tennessee River, and has an area of about 4100 square miles in Alabama, to which this particular type of country is chiefly confined. The greater part of the area is underlaid by horizontal strata of Mississippian (Subcarboniferous) age, but at the southeastern edge there is a long straight narrow valley of folded and faulted Ordovician and Silurian rocks, very similar to the Coosa valley described farther on. Much of the rock is limestone, and the soils are prevailingly reddish. The central part of the area is comparatively level, but near its edges are many ridges and rocky slopes. The greater part is now covered by soil surveys. The leading soil series are the Clarksville, Decatur, Huntington and Hagerstown, and the texture classes silt loam, stony and gravelly loam, clay loam, loam, rock, silty clay loam, and fine sandy loam. Evidently there is not much contrast in soil texture between this region and the preceding, but the vegetation and agriculture seem to indicate considerable chemical difference.

About 16 per cent of the original forest was evergreen. The commonest trees at present seem to be cedar, sweet gum, loblolly pine, red oak, scaly-bark hickory, sycamore, willow and beech. About 30 per cent of the area was "improved" in 1880, and the average expenditure for fertilizer in 1909-10 was about 25 cents an acre.

3. The plateau region, a continuation of the Cumberland plateau of Tennessee and Kentucky, covers about 3100 square miles in Alabama, where most of it goes by the name of Sand Mountain. Erosion has isolated several mesa-like outliers, of which Lookout Mountain is the largest. The rocks are nearly all sandstone of the lowest Coal Measures, and the strata are essentially horizontal. The surface is comparatively level for a mountain region, but stands several hundred feet higher than the valleys on either side, making it somewhat difficult of access. Most of the area has been covered by soil surveys, and about 98 per cent of the soils, not counting rock outcrop, are referred to the DeKalb series (named for DeKalb County, Alabama). The prevailing soil classes, are fine sandy loam, sandy loam, rock, stony loam, loam, and silt loam. There is more fine sandy loam here than in any other of the highland regions.

About 45 per cent of the forest is evergreen. The commonest trees are loblolly pine, short-leaf pine, post oak, red maple, poplar, sweet gum, cliff pine, white oak, chestnut oak and dogwood. Only about 8 per cent of the land was cultivated in 1880, and in 1909-1910 the farmers spent about a dollar an acre for fertilizers; all of which indicates soils below the average in fertility.

4. The coal basin region covers about 3300 square miles, and does not extend into any of the neighboring states. The rocks are a little younger than those of the plateau, more shaly, and more folded and faulted in some places. The topography is more dissected, mainly on account of the rocks being softer. The prevailing soil series are DeKalb and Hanceville, and the

texture classes silt loam, shale loam, fine sandy loam, very fine sandy loam and stony loam. Shale loam is more abundant here than in any other part of the state.

About 40 per cent of the trees are evergreen. The commonest species are loblolly pine, cliff pine, short-leaf pine, beech, long-leaf pine, red oak, white oak, sweet gum, red maple, post oak, poplar and dogwood. Only about 10 per cent of the area was cultivated in 1880, and even now coal mining employs about as many people as farming. The expenditure for fertilizers in 1909-1910 was about 75 cents an acre.

5. The Coosa valley, the southwestern extremity of the great Appalachian valley, which is over 1000 miles long, includes about 4000 square miles in Alabama, more or less interrupted by outliers of the neighboring mountain regions. The rocks are all Paleozoic, mostly older than Carboniferous, and nearly everywhere folded and faulted, making many narrow longitudinal ridges and minor valleys. Both limestone and sandstone are common. The principal soil series as mapped are Clarksville, Decatur, Huntington, Fort Payne,⁶ and Hagerstown, and there are at least a dozen others. Gravelly, stony and shale loams make up nearly half the total, and next in importance are loam, silt loam, clay loam, clay, and fine sandy loam. This region leads all the rest in stony loam and loam, and all the other highland regions in clay.

Evergreens make up about 45 per cent of the forest. The commonest trees seem to be loblolly, long-leaf, and short-leaf pines, sweet gum, post oak, red oak, black-jack oak, poplar, red maple, white oak, water oak and willow oak. About 20 per cent of the land was improved in 1880, and 64 cents an acre was spent for fertilizers in 1909-1910.

6. The Blue Ridge is represented in Alabama by a narrow strip trending northeast-southwest, and a few outlying ridges and peaks, probably not over 400 square miles in all. The rocks are pretty old and highly metamorphosed, and mostly siliceous. The topography is typically mountainous, and we find here the highest mountain in the state, Cheaha, 2400 feet above sea-level. Most of the area is covered by soil surveys, and the soils are referred chiefly to the Talladega⁷ and DeKalb series. The leading texture classes are slate loam, rough stony land, gravelly and stony loam, and silt loam. The first two are relatively more abundant here than in any other part of the state.

About half the trees are evergreen, and the commonest ones are long-leaf pine, short-leaf pine, black-jack oak, black oak, loblolly pine and chestnut oak. The area is too narrow for census statistics, but there is very little agriculture, on account of the rough topography.

⁶ Named for Fort Payne, Alabama, which is in this region; but most of the original Fort Payne soils in Alabama are now referred to the Clarksville series.

⁷ This is one of the few cases where the soil series name corresponds with the geological formation name.

7. The Piedmont region covers about 5000 square miles in Alabama, and extends northeastward to Pennsylvania. Its rocks are metamorphic or crystalline, and largely granitic. The topography is everywhere hilly, and there is running water on practically every square mile. About half the soils are referred to the Cecil series, and next in importance are the Louisa, Congaree and Talladega. The leading types are sandy loam, stony loam, slate loam, clay loam, loam, stony and gravelly sandy loam, and fine sandy loam. Some shade of red is the prevailing color, and minute flakes of mica are very characteristic. There is more sandy loam and more swamp than in any of the regions previously described.

About 45 per cent of the present forest is evergreen. The commonest trees seem to be long-leaf, loblolly and short leaf pines, sweet gum, black-jack oak, poplar, red oak, post oak, red maple, bay and dogwood. About 24 per cent of the acreage was cultivated in 1880, and thirty years later the farmers were spending 95 cents an acre annually for fertilizers.

8. The innermost division of the coastal plain in Alabama is the short-leaf pine belt. Including the Eutaw belt (which was separated before) it covers about 6600 square miles in Alabama, and extends northwestward across Mississippi into Tennessee. It is underlaid by clayey non-calcareous Cretaceous strata of the Tuscaloosa and Eutaw formations, and is moderately hilly and well watered. At least 16 different soil surveys touch this region, and about twenty soil series have been distinguished, of which the principal ones are the Orangeburg, Guin, Norfolk and Ruston. The leading texture classes are fine sandy loam, sandy loam, gravelly sandy loam, loam and gravelly loam.

About half the trees are evergreen. The commonest are loblolly, short-leaf, and long-leaf pines, sweet gum, bay, water oak, poplar, beech, willow, post oak, white oak, red maple, dogwood, willow oak and cypress. The soils seem to be a little below the average in fertility, for only 16 per cent of the area was "improved" in 1880, and the expenditure for fertilizers in 1909-1910 averaged 75 cents an acre.

9. The central long-leaf pine hills, about 850 square miles in extent, are surrounded by the short-leaf pine belt, and might be treated as a high and dry extreme of it. The underlying strata are of the Tuscaloosa formation, and composed of clay, sand and pebbles in varying proportions. It is a high and rather hilly region, almost mountainous in places, with many creeks and branches. It has been completely covered by soil surveys, but one of those is so old that it does not give much detail. The soil series recognized are the Orangeburg, Susquehanna, Norfolk, Guin, Ruston, and one or two others, and the leading types are sandy loam, gravelly sandy loam, gravelly loam, loamy sand and fine sandy loam. The first four are more prevalent here than in any other part of the coastal plain in Alabama.

Evergreens make up about 60 per cent of the forest. The commonest trees

are long-leaf pine, loblolly pine, black-jack oak (two kinds), short-leaf pine, turkey or upland willow oak, bay, red oak, red maple, poplar, and dogwood. The region does not cover as much as half of any one county, so that no agricultural statistics are available, but probably not over 15 per cent of it is under cultivation even yet, on account of the rough topography and rather poor soil.

10. The black belt or central prairie region, covering about 4300 square miles in Alabama, is one of the most distinct and sharply defined divisions of the coastal plain. It has no counterpart farther east, but extends northwestward across Mississippi, and reappears in Texas. It coincides almost exactly with the area of the Selma Chalk or Rotten Limestone, one of the Cretaceous formations. Most of the soil is some shade of gray. The topography is undulating, not very hilly, and water is rather scarce, as in many other fertile and calcareous regions. The summers are drier here than in any other part of the coastal plain of Alabama, a fact which may have some bearing on the soil conditions.⁸

The soils have been referred to about twenty series, of which the most extensive are the Houston,⁹ Norfolk, Orangeburg, Susquehanna, Oktibbeha, Waverly, Yazoo, Ocklocknee, and Trinity. Over half the soil is classed as clay, and over one-fifth as fine sandy loam. The only other type making over 3 per cent of the total is loam.

A part of this region, possibly 10 per cent, was treeless when first discovered. About 30 per cent of the trees in the remaining forests are evergreen, but this figure decreases northwestward, with the drier summers. The commonest species seem to be loblolly pine, sweet gum, post oak, short-leaf pine, long-leaf pine, red oak, hackberry, willow oak, cedar, cottonwood, willow, elm and sycamore. About 45 per cent was cultivated in 1880, and only 35 cents an acre was spent for fertilizers in 1909-1910, indicating soils above the average in fertility, for the coastal plain especially.

11. The Chunnenuggee Ridge, or blue marl region, is chiefly confined to Alabama, where it covers about 2300 square miles. It is underlaid by marly strata of the Ripley formation (Upper Cretaceous), and has a moderately hilly topography, with more streams than the black belt. The soil series are Norfolk, Susquehanna, Orangeburg, Ruston, Kalmia, and nine or ten others, and the principal texture classes are sandy loam, fine sandy loam, sand, clay, fine sand, meadow, etc.

⁸ See Geol. Surv. Ala., Monog. 8, p. 19, 24 (footnotes). 1913.

⁹ This series name was first used in the reports on Perry County, Alabama, and the "Brazoria area," Texas, about 14 years ago, and is apparently derived from Houston, Texas, which is in a region quite different geologically and topographically from the black belt of Alabama and Mississippi (which however does have a counterpart in the interior of Texas). If it should ever seem desirable to separate the soils of the undulating Cretaceous black belt from those of the level Neocene coast prairie an appropriate name would be Noxubee, which is the name of a large creek in the black belt in Alabama and Mississippi and of a county in Mississippi.

About 45 per cent of the trees are evergreen, and the commonest species are loblolly, long-leaf and short-leaf pines, sweet gum, spruce pine, bay, post oak, red oak, water oak, poplar, and willow. This too is a more fertile region than the average of the coastal plain, and about 40 per cent of the area was classed as improved land in 1880. The expenditure for fertilizers in 1909-1910 was 79 cents an acre.

12. The post-oak flatwoods is a peculiar region almost confined to Alabama and Mississippi, and covering about 335 square miles in Alabama. It is underlaid by one of the oldest of the Eocene formations, a grayish non-calcareous clay at the surface, passing downward, in some places at least, into a sort of black shale. The topography is comparatively level, as the name implies, and streams are scarce. Only one of the Alabama soil surveys touches this region, and that (Sumter Co.) was published about 12 years ago. But the adjoining county of Noxubee in Mississippi, surveyed six years later, has been combined with it here to make the statistics more accurate. The prevailing soil series are the Lufkin,¹⁰ Norfolk, Pheba,¹¹ Orangeburg, and Waverly, and the texture classes fine sandy loam, clay, clay loam, loam, sandy loam and silt loam. There seems to be proportionately more clay loam and loam here than in any other division of the coastal plain.

About 45 per cent of the trees are evergreen. The commonest are loblolly and short-leaf pine, post oak, sweet gum, red oak, water oak, willow oak and beech. The region is too narrow to apply census statistics to, but it is not cultivated much, on account of the "cold" stiff clayey soil and the scarcity of good water.

13. The southern red hills is one of the largest divisions of the coastal plain, extending from South Carolina to Mississippi and reappearing farther west. It covers about 8000 square miles in Alabama. The underlying rocks are Eocene, but most of the surface material is a reddish sandy clay which may be of much later age. The topography varies from nearly level to hilly and even mountainous. (The "mountains" extend from Monroe County northwestward into Mississippi, and are high enough to be pierced by a railroad tunnel in each state, apparently the only tunnels in the whole coastal plain.) Streams are common, most of them bordered by swamps. About fifteen soil series have been recognized, of which the principal ones are the Orangeburg, Norfolk, Susquehanna, Greenville,¹² Kalmia, and Ruston. By texture the soils are classed as fine sandy loam, sand, sandy loam, fine sand, swamp, clay, etc.

Evergreens are estimated to make 58 per cent of the forest. The commonest trees are long-leaf, loblolly and short-leaf pines, sweet gum, spruce pine, red oak, water oak, bay, magnolia and poplar. About 17 per cent of the

¹⁰ Named for Lufkin, Texas.

¹¹ Named for Pheba, Mississippi, which is in this same flatwoods belt.

¹² Named for Greenville, Alabama, which is in this region.

area was cleared in 1880, and thirty years later the farmers were spending \$1.20 per acre annually for fertilizers.

14. The belt known as the lime hills extends from Wayne County, Mississippi, to Conecuh County, Alabama, and reappears in West Florida as the Marianna red lands. It covers about 1300 square miles in Alabama and 450 in Florida. The underlying rock is chiefly limestone of Upper Eocene age, but the surface is largely red loam. The topography is moderately hilly, with a few caves and natural bridges, but with plenty of streams and few or no ponds. The soil series are Orangeburg, Norfolk, Greenville, Ruston, Susquehanna, Ocklocknee, and seven or eight others, and the texture classes fine sandy loam, sandy loam, swamp, sand, coarse sandy loam, stony clay, etc. The last is more prevalent here than elsewhere in the coastal plain, and the same may be said of rock outcrop.

The proportion of evergreens has been estimated at 44 per cent in Alabama and 49 per cent in Florida. The commonest trees seem to be loblolly, longleaf, short-leaf and spruce pines, sweet gum, red oak, beech, magnolia, bay, dogwood, cedar, water oak, black-jack oak, white oak and cypress. No agricultural statistics are available, on account of the narrowness of the area, but agriculture has long been the leading industry.

15. The lime-sink region extends from Covington County, Alabama, and Walton County, Florida, northeastward about to the Savannah River, and covers about 1300 square miles in Alabama and 1600 in Florida. The underlying formation is limestone of the Vicksburg formation, more or less silicified and nearly everywhere covered with later clays and sands. The topography is comparatively level, and diversified by many shallow ponds and a few lime-sinks. Streams are not abundant, much of the drainage being subterranean. There are only two soil surveys for this region as yet (Covington County, Alabama, and the "Marianna area," Florida), and both of those include parts of two or more other regions, so that we are not in a position to give very accurate statistics of the soils. Over half are referred to the Norfolk series, and the rest are Tifton, Myatt, Kalmia, etc. Fine sandy loam, sandy loam, swamp, sand and fine sand make up about 99 per cent of the total.

About 70 per cent of the trees are evergreen. The commonest trees are long-leaf pine (about 45 per cent), pond cypress, slash pine, loblolly pine, forked-leaf black-jack oak, black gum, turkey oak, bay, red oak, sweet gum and magnolia. Very little of the land, possibly 3 per cent, was classed as "improved" in 1880, but by 1910 about 26 per cent was cultivated, with the aid of about \$1.40 worth of fertilizers per acre per year.

16. What may be called the southern pine hills extends from near the Savannah River in Georgia with some interruptions to Louisiana, and covers about 5000 square miles in Alabama and 3800 in West Florida. Within a

few hundred feet of the surface there are only unconsolidated non-calcareous sandy and clayey strata very similar to those of the central pine hills, but of much later age, probably not older than Miocene. The topography is hilly or undulating to flat, with running water on nearly every square mile, and many shallow ponds besides. Very few of the ponds are deep enough to hold water throughout the year.

The soils have been referred to about twenty series, of which the Norfolk covers over half the area. Next in importance are the Ruston, Orangeburg, Kalmia, Tifton, and Myatt. The principal texture classes are fine sandy loam, sand, sandy loam, fine sand, loamy sand and swamp.

The proportion of evergreens has been estimated at 74 per cent in Alabama and 76 per cent in Florida. There is more long-leaf pine than all other trees combined, and where undisturbed by lumbermen it forms beautiful open park-like forests, with very little underbrush. Other common trees, growing mainly in swamps and ponds or on sandy ridges, are slash pine, forked-leaf black-jack oak, loblolly pine, bay, pond cypress, turkey oak, and black gum.

The development of agriculture in this region has been rapid in recent decades. The improved land jumped from about 1 per cent in 1880 to 8 per cent in 1910. This increase would have been impossible without the aid of fertilizing materials from the mines of Florida, Germany, Chile, etc., the expenditure for which increased from 95 cents an acre in 1879-1880 to \$2.09 in Alabama and \$2.33 in Florida in 1909-1910.¹³

17. The Mobile delta comprises the swamps of the Mobile and Tensaw Rivers (continuations of the Tombigbee and Alabama) within the influence of tide-water, an area of about 300 square miles. This delta is long and narrow, and occupies what appears to be a "drowned valley" in the pine hills. Its lower extremity at present is just about opposite the city of Mobile, and is slowly moving seaward, but will hardly reach the mouth of Mobile Bay for several thousand years. The whole area is inundated almost every spring by muddy waters carrying the cream of the soil from an area of about 40,000 square miles in Georgia, Alabama and Mississippi, some of which is very fertile. The Apalachicola River has a similar but smaller delta, shown on the soil map of Franklin County, Florida, but it is mainly on the east side of the river, in Middle Florida, and has not been counted in the following statistics. The Mobile delta is nearly covered by the soil surveys of Baldwin, Mobile, and Clarke Counties, Alabama, in which nearly half its area is classed as Swamp, and the rest chiefly as "Ocklocknee¹⁴ clay" and "Ocklocknee silt loam."

¹³ For a discussion of the influence of imported fertilizers on agriculture in the pine lands see Jour. Geogr., vol. 15, p. 42-48. October 1916; and Geogr. Rev., vol. 2, p. 366-367. November, 1916.

¹⁴ This series name, derived from the Ocklocknee River of Florida, which rises among the pine hills of South Georgia (where it is spelled Ochlocknee) and is seldom muddy, does not seem very appropriate for the alluvium of large rivers that are always muddy.

About 13 per cent of the trees are evergreen. The explanation of the occurrence of any evergreens at all in such fertile soil is probably that lack of aeration limits the availability of the potassium compounds, and thus compels some of the trees to economize by keeping their leaves more than one season. The commonest trees seem to be black gum, cypress, bay (this is the commonest evergreen, chiefly toward the lower end of the delta, where the seasonal fluctuation of the water is at a minimum and therefore the aeration of the soil least), willow, sweet gum, red maple, and cottonwood. There is no agriculture except perhaps at the extreme edges of the swamps.

18. The Gulf coast strip is a narrow belt of islands, dunes and marshes along the coast from Middle Florida to Mississippi. About 65 per cent of its area is classed in the soil surveys as coastal beach, sand-hill, etc., and 27 per cent as tidal marsh. About 80 per cent of the trees are evergreen. The commonest are spruce pine (a different species from the one in the interior of the coastal plain), long-leaf-pine, slash pine, live oak (two species), forked-leaf black-jack oak, turkey oak and cedar. There is practically no agriculture.

19. The Knox Hill country is an area of about 50 square miles lying between the lime-sink region and the pine hills in Walton County, Florida. Its geology is not well known, but the underlying formations are believed to be at least in part Miocene. It is a region of reddish clayey hills and running water, strongly resembling some parts of the central short-leaf pine belt of Alabama. In some places the soil is so clayey that the roads have to be surfaced with sand or poles, which is very unusual in Florida. There is no soil survey for this region yet, but much of the soil would probably be called "Orangeburg fine sandy loam" or something of the sort.

A majority of the trees are evergreen, and the commonest species are loblolly pine, short-leaf pine, red oak, long-leaf pine, dogwood, magnolia, sweet gum, holly, spruce pine, beech, white oak, bay, black-jack oak and black gum. Agriculture has been an important industry here for nearly a century. At present probably not more than 25 per cent of the area is under cultivation, but the cultivated acreage is said to have been larger before the Civil War, when commercial fertilizers were unknown, and farming in Florida was confined to the few areas of naturally fertile soil.¹⁵

20. Holmes Valley is an area of 25 or 30 square miles lying between the lime-sink region and that next to be described, in Washington County, Florida. It resembles the Knox Hill country in being a red hill region surrounded by sandy pine lands, but differs in having less running water and coarser and less clayey soils. The soil is evidently quite fertile, however, for most of it is or has been cultivated; and in the sixth volume of the Tenth Census (page 225) Dr. Smith cites a case where a tract of land in this region was producing nearly a bale of cotton to the acre in 1880 after being culti-

¹⁵ See Geogr. Rev., vol. 2, p. 363. November 1916.

vated for thirty-five years, without fertilizers. From the abundance of sweet gum, which is common in some known phosphatic soils, it is reasonable to suppose that the soil of this region is well supplied with phosphorus, though neither physical nor chemical analyses are available yet.

Evergreens are less abundant here than in the preceding region, and perhaps not in the majority. The commonest trees seem to be loblolly pine, sweet gum, long-leaf pine, black gum, dogwood, bay, beech, water oak, white oak, magnolia, red oak, post oak and hickory.

21. The West Florida lake region has an area of perhaps 350 square miles. Nothing definite is known about its geology, the surface being almost completely covered with sand. The surface is comparatively level except for being pitted with numerous approximately circular depressions from several to a few hundred acres in extent, most of them containing lakes which never dry up.

A part of the region is included in the soil survey of the "Marianna area," and the prevailing soil is classed as "Norfolk sand." About 75 per cent of the trees are evergreen, and long-leaf pine and forked-leaf black-jack oak constitute the bulk of the forest. No census statistics are available, but there is very little agriculture yet, on account of the very sandy soil.

22. The Apalachicola flatwoods region extends from Bay County to Wakulla County, Florida, just back of the coast strip, and about 1200 square miles of it lies in West Florida. No geological formations older than Pleistocene are known here, and the soil is nearly all sand, where it is not covered by peat, muck or something else. The surface is nearly everywhere flat and damp. The soil survey of Franklin County, Florida (published last fall), covers part of this region, and although that county is mostly in Middle Florida, there is no reason to suppose that its soils are not equally typical of the West Florida portion.

The principal soil series are Hyde, Plummer, Norfolk, Portsmouth, and Leon, and the texture classes fine sand, swamp, loamy fine sand, etc. There is more fine sand here than in any region previously described. About 75 per cent of the trees are evergreen, but this figure would be higher if the Apalachicola River swamps were excluded. The commonest trees are long-leaf pine, pond cypress, slash pine, black pine, bay, black gum, sweet gum and red maple. Less than 3 per cent of the area is under cultivation. The expenditure for fertilizers in Franklin County in 1909-1910 was about a dollar an acre, but it would be much greater if anything more than a few of the richest spots were cultivated.

STATISTICS

The following tables show the percentage of each type of soil in each of the regions above described, except some of the smallest ones, for which no statistics are available. One table is for the highlands and one for the coastal

plain, and the averages are given for each of these major divisions as a whole. In each table the highest figure in each row is printed in heavier type, to show what classes of soil are more abundant in a given region than in any other. The percentages are given only to the nearest integer, to save space and also to avoid making them appear more accurate than the facts warrant. Percentages below 0.5 are therefore represented by 0. The soil classes are arranged approximately in order of texture, the coarsest first.

At the bottom of each table the statistics of evergreens, improved land, and fertilizers already given are brought together, and also summarized for the two major divisions.

TABLE 1
Soils of highlands in Alabama

SOIL CLASSES	REGIONS							Whole highlands
	1. Barrens	2. Tens. valley	3. Plateau	4. Coal basin	5. Coosa valley	6. Blue Ridge	7. Piedmont	
Rock outcrop, etc.....		9	10	5	2	23	0	6
Fine sand.....		0						0
Stony, gravelly or chert loam.....	36	18	8	7	46	19	20	19
Shale or slate loam.....			4	27	2	54	14	11
Stony or gravelly sandy loam.....			2	2	2		9	3
Stony silt loam.....			1					0
Coarse sandy loam.....							1	0
Sandy loam.....		0	19		1	0	24	10
Fine sandy loam.....		6	39	15	5	0	5	13
Very fine sandy loam.....				10				1
Loam.....		12	7	0	15		10	8
Silt loam.....	44	28	5	35	10	3		15
Silty clay loam.....	1	7						1
Clay loam.....	18	18			9	1	11	8
Stony clay.....		0			0		2	1
Clay.....	0	1	3		6		0	2
Swamp or meadow.....		1	1		1		2	1
Percentage of evergreens.....	30	16	45	40	45	50	45	39
Percentage of improved land, 1880.....	?	30	8	10	20	0	24	21
Expenditure for fertilizers per acre, 1909— 10 (dollars)	?	0.25	1.00	0.75	0.64	?	0.95	0.63

Some comments on these tables will be of interest. It is, of course, not at all surprising that rock outcrops should be far more extensive in the highlands than in the coastal plain, for the strata of the latter, being much younger, have never been subjected to enough pressure from overlying materials to harden them into rock. What few rocks do occur in the coastal plain are chiefly the result of cementation of sand by iron oxide, or of deposition of

calcium carbonate by marine organisms. The only rock outcrop in the whole coastal plain of the United States reported in Bulletin 96 of the Bureau of Soils (page 301) is 384 acres of limestone in the "Marianna area," Florida, and this forms the basis of the first figure in column 14. But the "Lauder-

TABLE 2
Soils of coastal plain in Alabama and West Florida

SOIL CLASSES	REGIONS												Whole coastal plain
	8. Short-leaf pine	9. Central pine hills	10. Black belt	11. Chunnenanegree	12. Post oak	13. Red hills	14. Lime hills	15. Lime-sink	16. So. pine hills	17. Mobile delta	18. Coast strip	22. Apalachicola	
Rock outcrop.....						0							0
Gravelly sand.....					1				2				0
Coarse sand.....	3			3		1	2	0	0				1
Sand, sand-hill, coastal beach.....	3	2	2	17	2	21	9	12	23	65			12
Fine sand.....	0	1		7		8	2	9	8	0	82		6
Loamy sand (fine or coarse).....		15		0		3	0		7		1	4	3
Stony or gravelly loam.....	5	19		1		0							2
Stony or gravelly sandy loam.....	10	21		3		1	2		2				4
Coarse sandy loam.....	0			0		0	3	0					0
Sandy loam.....	21	33	2	24	4	18	14	21	14				16
Fine sandy loam.....	37	7	22	23	35	37	45	45	34	1			32
Very fine sandy loam.....	0			5									0
Loam.....	5		6		9	1	2		2	3			3
Silt loam.....	4				4	0	2		1	17			2
Silty clay loam.....	0			0									0
Clay loam.....	1		2		9	0	1				3		1
Stony clay.....						1	3						0
Clay (including black clay).....	4		60	11	33	4	2		1	28			11
Swamp, meadow, muck, etc.	5	4	2	6	3	5	13	13	6	47	7	14	6
Tidal marsh.....										127			0
Percentages of evergreens ..	50	60	30	45	45	58	47	70	75	13	80	75	59
Improved land, 1880.....	16	?	45	40	?	17	?	?	1	0	0	1	14
Expenditure for fertilizers per acre, 1909-10 (dollars).....	0.75	?	0.35	0.79	?	1.20	?	1.40	2.18	0	?	1.04	0.93

dale stony clay" in the southern part of Clarke County, Alabama (not that in the northern part) is practically the same thing, and belongs to the same region; and there are many rock outcrops too small to map in other divisions of the coastal plain.

Pure sand as a soil type is almost wanting in the highlands, but fairly common in the coastal plain. The various stony loams, silt loams, and clay loams are common in the highlands, but scarce south of the central pine belt, while the sandy loams are naturally commonest southward. There is more pure clay soil in the coastal plain than in the highlands, a fact not easily explained in a few words.

It has long been known that swamps are more extensive in the coastal plain than in the interior, but that fact has perhaps never been shown statistically before. Although the percentages for swamps cannot be regarded as very accurate, for reasons given on an earlier page, it is unquestionable that they are several times as prevalent in the coastal plain as in the highlands; a fact doubtless correlated with the "immaturity" of the topography, and also with the heavier summer rainfall toward the coast, which counterbalances the evaporation to a considerable extent.

The evergreen percentages range from 13 in the Mobile delta to 80 in the coast strip, and average about 52 for the whole area mapped. The highest percentage of improved land is in the black belt, and the highest expenditure for fertilizer in the southern pine hills.

The correlations between evergreens, improved land and fertilizers are very significant, especially if we disregard those regions where agriculture is undeveloped on account of rough topography or periodical inundation, or better still, consider only the two major divisions, thus eliminating local irregularities. The highlands, having richer and finer soil on the average than the coastal plain, have relatively more deciduous trees and more improved land, and spend less for fertilizers. The difference in the amount of improved land is less now than it was in 1880, however, for the increasing consumption of fertilizers tends to obliterate original differences in soil fertility.

A SOIL SAMPLER FOR BACTERIOLOGICAL AND CHEMICAL PURPOSES

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Samples of soil for bacteriological examination are generally obtained by digging a trench with a spade, sections being then taken with a sterilized spatula from the vertical wall at the end of the trench. This involves a considerable amount of labor and inconvenience, especially when sub-surface portions are desired. Furthermore, on plots planted to crops, the trench method disturbs the soil mass in an objectionable way, particularly when samples are taken at frequent intervals. Thus when composite samples are obtained to ensure a more uniform sampling and when these are taken six or twelve times a year, a method causing much digging over of the soil is not feasible.

For chemical and physical studies soil augers may often be used, the procedure being to enlarge and clean the boring with the larger auger and sample with the smaller. But this method is not satisfactory for bacteriological studies as there is always more or less contamination of the lower sections by the soil from the surface falling down into the boring. Moreover, it is impossible to remove the soil from the auger without exposing and contaminating it. Bacteriological samples may be conveniently obtained from the surface layer only, using the apparatus described by Noyes in the Journal of the American Society of Agronomy (vol. 7, no. 5, p. 221).

THE SAMPLER

In order to obtain samples below the surface the author devised a sampler, the five parts of which are illustrated by longitudinal drawings in figure 1. C shows the surface shield, the upper flange, *a*, being 10 inches in diameter and $\frac{1}{2}$ inch thick while the tube, *b*, is 4 inches long and has an inside diameter of $2\frac{1}{4}$ inches.

The sampling tube, *A*, is $3\frac{1}{2}$ feet long and has an inside diameter of $1\frac{1}{4}$ inches. It is divided into two parts, *a* and *b*. Piece *a* (shown by *B*) is 11 inches long and has a point made so that the core slides easily up through the tube. The inner shoulder above the cutting edge is about $\frac{1}{8}$ inch wide. Piece *a* is attached to piece *b* by means of a bayonet, or groove and key joint, made so that it closes and tightens when the upper part of the tube is turned to the right. A slight twist to the left enables one to detach the lower

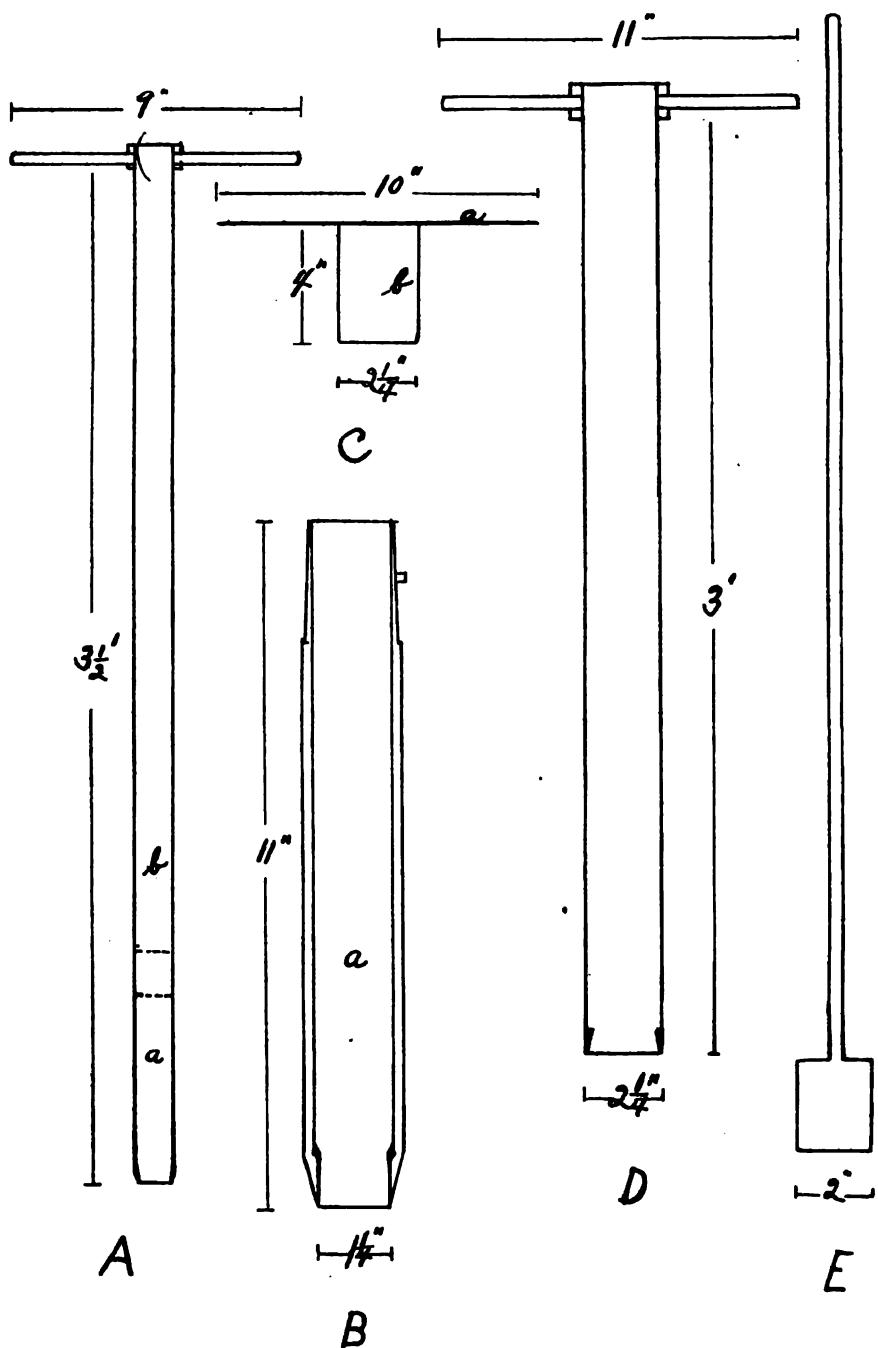


FIG. 1. LONGITUDINAL SECTIONS OF SOIL SAMPLER.

A, The sampling tube; *B*, lower end of the sampling tube of *A*; *C*, the surface shield; *D*, the cleaning and enlarging tube; *E*, tamper to be used with cleaning tube when necessary.

piece and remove the core of soil. The shoulders of this joint taper slightly into each other so as to come together and take up any looseness resulting from wear.

Instead of the bayonet joint an ordinary pipe coupling union may be used, although it is not as handy or as easy to separate and to unite the two parts of the tube when this device is employed.

The cleaning tube, *D*, has an outside diameter that permits it to slip easily but snugly through the surface shield, *C*. Otherwise, the construction is similar to that of the sampling tube except that the point does not taper in to obtain the cutting edge. This should be avoided in order that particles of soil may not work up between the cleaning tube and the surface shield in the process of sampling. Each of the tubes is reinforced at the top with a flange $\frac{1}{16}$ inch thick and the handles are screwed through the flange and the wall of the tube. The tubes may be laid off into the desired divisions, for instance every 6 inches, by cutting shallow grooves in them with a lathe.

In loose dry soil it is advisable to use the tamper, *E*, to tamp down the soil in the cleaning tube before removing it from the boring, thus making sure that the bottom of the boring is well cleaned and that no soil drops back into it. In moist soils this precaution is hardly necessary.

The material used throughout consists of a good grade of tool steel, with the exception of wooden handles on the larger tube and the cylindrical wooden plunger of the tamper.

METHOD OF SAMPLING

To use the sampler the surface shield is first thrust into the ground. This prevents any of the loose surface soil from falling into the boring as it is made. The first or surface section is then taken with the sampling tube (plate I, fig. 1). Only the small lower piece *a* (fig. 1, *A*) needs to be sterile. It may be plugged with cotton and sterilized in a hot-air oven before taking it from the laboratory. For sterilization in the field alcohol is used. Contamination from above is prevented by inserting a cotton plug into the bottom of piece *b* (fig. 1, *B*) and then flaming it. When a core of the desired length is in the sampler, piece *a* is removed by a slight turn to the left and the core is allowed to slide out through the upper end into a sterile glass jar. In case one does not desire to disturb the core, the tube may be capped and similar tubes, previously sterilized, used for the other sections. To obtain the next section below the surface the cleaning tube is inserted, to clean and enlarge the boring down to the desired depth, using the tamper (fig. 1, *E*) to compact this core in case it is too loose to remove completely without tamping. The sampling tube is then lowered into the enlarged boring, care being taken that its point does not touch the side walls. In this manner the desired number of sub-surface samples may be obtained. If composite samples are desired additional borings may be made over a given area, sections from a given depth being added to the jar marked to receive them.

In sampling for chemical analysis the procedure is similar except that the apparatus need not be sterilized.

The sampler fits together conveniently and may be carried by means of a strap (plate I, fig. 2). Its total weight is about 12 pounds. A mechanic of ordinary ability should have no trouble in making one. It is giving satisfactory service in bacteriological investigations which are being conducted at the New Jersey Agricultural Experiment Station.

An expression of appreciation is extended to Dr. J. G. Lipman and Prof. A. W. Blair for their kindly interest and constructive criticism.

PLATE I

FIG. 1. Apparatus in position for taking the surface section. The arrow points to the joint in the sampling tube.

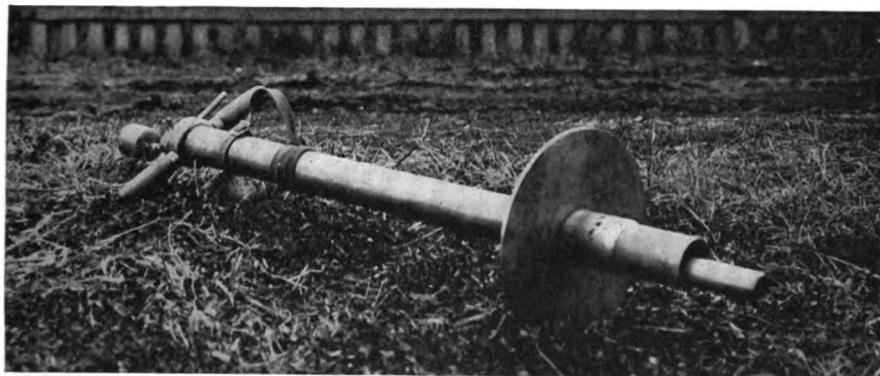
FIG. 2. Showing the relative size of the tubes.

SOIL SAMPLER FOR BACTERIOLOGICAL PURPOSES
J. R. NELLER

PLATE 1



1



2

EXPERIMENTS IN METHODS FOR DETERMINING THE REACTION OF SOILS

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INTRODUCTION

In former papers (6, 7, 9, 10), the author has published results obtained from extensive investigations in determining the influence the reaction and basicity of the soil exerts upon its condition. He was able to state that this influence is very complicated and many-sided. He demonstrated that by determining the above-named characteristics of the soil, comparatively certain information as to the need for lime in the soil might be obtained. This is of especial interest for practical purposes.

The methods used in the investigations mentioned above were largely qualitative. Despite this fact they have given reliable information as to the need for lime in the soil. The author, as indicated (9, p. 382), thinks that this is due to the fact that the need for lime in soil is not (as in the case of need for nitrogen, phosphoric acid or potassium), first of all determined by the amount of nutritive elements present in the soil in a form available for plants, but expresses a particular condition of the soil,—that is, *the presence or absence of certain basic substances*.

The nature of the question is such, that methods for an accurate quantitative determination of the above-named characteristics of soils might in many instances give much interesting information. For the present we will limit ourselves to investigations of the need for lime. We find that by using the qualitative methods (acid, litmus and *Azotobacter* tests), known at present, we can determine whether or not the soil contains the necessary basic substances, but we can obtain only faint suggestions as to the degree of this need, and the supply of these substances.

We must be careful not to overexaggerate the practical importance which a quantitative determination of the acidity or basicity of the soil might have in indicating its lime-requirement. We must remember that by treating lime-poor soils with chalk or marl, an attempt is made to give a surplus of lime sufficient for a number of years—in other words, a surplus of basic substances; and that the more complete and expensive the experimental methods, the less the possibility for using them in practical work. Yet the information obtained by using such quantitative methods will doubtless give far greater

insight into the complicated relationship between soil and lime, and will be of very special importance in research work on the influence exerted by lime, applied in varying amounts, upon the condition and productivity of various soils.

I need merely mention the importance of the qualitative determination method in special research work on the influence of the acidity or alkalinity of the soil on its physical, chemical and biological conditions.

During the past year the author has investigated one phase of the above-mentioned problem, i.e., determining quantitatively the acidity of the soil; and the results of the investigations brought to a temporary close are now submitted in the first main division of this paper.

In the second main division is a series of comparative investigations of methods for making a qualitative determination of the reaction of the soil, and in connection with these some quantitative determinations of the ability of the soil to free acids from various salts.

Probably at some future time (and perhaps starting from other principles), I may have an opportunity for a renewed study of the entire complicated problem of the determination of the reaction of soils. The results here presented are but a modest contribution toward the solution of that problem.

I. QUANTITATIVE DETERMINATION OF THE REACTION OF THE SOIL

1. *Methods for making a quantitative determination of the acidity of the soil*

The method hitherto used in the majority of cases for determining the degree of acidity in the soil, is the one recommended by B. Tacke (29), leader of the Moor-Experiment Station in Bremen. The method is particularly adapted to the investigation of humus-soils.

The principle underlying the method is to determine the amount of CO₂, which a certain quantity of soil mixed with a surplus of calcium carbonate is able to free from this salt.

As Tacke himself indicates, this method does not give absolutely constant or accurate results. The carbonic acid never wholly ceases to generate from the humus mixed with carbonates, a condition believed to be occasioned by "inner reactions" of humus stuffs taking place during the treatment. In order to strengthen this weakness in the method Tacke's collaborator, H. Süchting (26, 27), has modified the method so that instead of making a direct determination of the carbonic acid freed by the action of soil acids on carbonates, a determination is made of the amount of carbonates left in the retort after reaction has taken place. A surplus of hydrochloric acid is added to this remainder and the amount of carbonic acid generated is determined in the usual way. The difference between the amount of carbonic acid added, and the amount of carbonic acid remaining in combination, gives us an expression for the acid content of the soil.

Süchting shows that this modification of Tacke's method gives greater surety and accuracy than the original method, for the "inner reactions" of humus stuffs referred to above are here present in but very limited amounts. In both methods the reaction takes place at ordinary room-temperature.

In 1903, three Americans, Hopkins, Knox, and Pettit (17), published a method for determining the acidity of soils based upon the ability which acid soils inter-reacting upon neutral salts have to free acids from them. In 1905 the American agricultural chemists convened in Washington adopted this method as the one to be used for the present when determining the acidity of the soil. The method proposed is as follows.

One hundred grams of soil are placed in a 400 cc. retort and covered with so much of a 5 per cent sodium chloride solution, that it, together with the water contained in the soil, yields 250 cc. of liquid. The retort is placed in an agitator and shaken for three hours (or shaken by hand every half-hour during a period of twelve hours). The mixture is filtered and the acid content of 125 cc. of the filtrate determined by titration with diluted NaOH, using phenolphthalein as an indicator.

If for the 125 cc. of liquid taken from the retort an equal amount of a fresh sodium chloride solution is substituted and the above process followed, we find, according to investigations made by Hopkins and his collaborators that the next 125 cc. of liquid titrated contains about two-thirds as much acid as the first portion, and that portion no. 3, contains two-thirds as much as portion no. 2, etc. From this constancy we are able to compute the factor with which the result of the first three titrations may be multiplied in order to express the absolute acidity. This factor is estimated to be about 3.¹

Based on the determinations made according to this method, and the determinations as to the weight of soil per cubic measure, it is possible to estimate how large an amount of lime will be necessary for neutralizing an acre of soil to a certain depth, and the above-mentioned scientists take for granted that this amount expresses the lime-requirements of the soil.

As Baumann and Gully (4) have later proved, the ability of the soil to free acid from salts of so strong an acid as hydrochloric acid is very limited, and these scientists therefore emphasize the fact that when it is a question of an approximately absolute determination of the ability of the soil to absorb bases, it is preferable to use salts of weak acids. Veitch (32) has pointed out that the deduction made by Hopkins and his collaborators that a combination of sodium chloride solution with acid soil frees hydrochloric acid is incorrect, and that the acid reaction of the liquid is due to the formation of aluminum chloride.

A similar method has been proposed very recently by the Japanese scientist G. Daikuhara (12) who, however, instead of using sodium chloride solution, used a 1/1 N solution of potassium chloride, for he points out the fact that the soil is able to absorb a far greater amount of potassium than of

¹ Later Hopkins (17), as a result of continued experiments, prefers to use factor 4.

sodium. Using this solution the approximately absolute acidity of the soil may be computed by multiplying the first result of titration (see above) by the factor 3.

It is especially interesting to note that Daikuhara confirms Veitch's statement, that by a reaction between acid mineral soil and a potassium chloride solution, aluminum (and in certain instances iron) compounds will become soluble and too, that *the amount of these substances corresponds very closely to the amount of alkalies used up in titration*. Daikuhara therefore concludes that the acidity of mineral soils depends upon certain aluminum and iron compounds showing an acid reaction for litmus and which are absorbed by the soil colloids. The above-mentioned compounds are insoluble in water. By treating neutral soils with aluminum or iron chlorides, and then washing them out, the soils were found to cause a strong acid reaction in a potassium chloride solution.

To prove the correctness of this method for determining the acid content of soils, Daikuhara has mixed three different acid soils with just that amount of carbonate of lime, which investigation has found necessary for neutralizing these soils. After the mixture had stood for a longer time, the acidity of the soil was again determined. The result showed that the soil specimens became just neutral upon the addition of the computed amount of carbonate of lime. Furthermore, the author has arranged experiments in tubs with various soils and various plants, and he claims he has proved that the amount of lime necessary to neutralize the acidity of the soil corresponds very closely with the amount most beneficial to the plants. A more detailed report of these experiments is promised.

By means of the litmus test, and by examining the effect of a neutral solution of potassium chloride on the soil, Daikuhara has shown, that a very great quantity of acid mineral soils are to be found in Japan and Korea.

The same year as Hopkins and his collaborators published their method, another American, Veitch (31, 32), proposed using lime-water in determining quantitatively the acidity of the soil. His method in brief is as follows.

Of the given acid soil weigh off three equally large portions. To these apply various amounts of the standard solution of lime—for instance, to the first portion 10 cc., to the second 20 cc., and to the third 30 cc. Vaporize over a water-bath, and wash the vaporization remainder with the help of 100 cc. of distilled water into a Jena retort. The mixture should stand (shaken from time to time) until the next day. Then 50 cc. of the transparent liquid over the precipitate (or of the filtrate) should be placed in a Jena beaker, a few drops of phenolphthalein solution added, and the liquid vaporized until it assumes a reddish color, or if this color does not appear, until it has a volume of 5 cc.

After having roughly determined, by this method, how much lime is necessary to neutralize the soil, a more exact determination of its acidity is made, and by varying the amount of lime-water added within narrow limits (for instance, 2 cc.) an attempt is made to determine the smallest amount of lime-

water, which, when used according to the method described above, will bring about the characteristic red color reaction for phenolphthalein.

The method is very detailed, requires much time and is therefore difficult to use when many determinations of the acidity of soils are to be made. Besides, it is not easy to determine the saturation-point with sufficient accuracy, and finally, warming the soil with an alkaline solution is a weakness in the method (see further p. 120).

Yet Veitch's method is very generally used by American soil scientists, and the acidity of the soil, or what is considered the same—the lime-requirement of the soil, is expressed by the amount of lime which a weight unit of soil may absorb, or when the results are to be used by practical agriculturists—by that amount of lime which must be applied per acre in order to neutralize the top layer of soil.

Quite similar to Veitch's method is one recently published by H. B. Hutchinson and K. MacLennan (18). Instead of calcic hydrate, a 1/50 N solution of bicarbonate of lime is used. The authors describe their method as follows.

To determine the acidity of the soil or its lime-requirement, place 10 to 20 grams of soil in a retort of 500 to 1000 cc. capacity, with 200 to 300 cc. of a *ca.* 1/50 N calcium bicarbonate solution. Remove the air in the retort by passing a carbon dioxide stream through it, thus making sure that no calcium carbonate will be precipitated during the analysis. Place the retort in an agitator and shake for three hours. Filter the liquid. Of the filtrate take an amount equal to half the amount of calcium-bicarbonate solution originally present. Titrate this with a 1/10 N acid, using methyl orange as an indicator. The difference between this result and the result obtained from titrating the liquid in its original condition, indicates the amount of lime absorbed. One cc. of 1/10 N acid corresponds to 5 mgm. of calcium carbonate.

The method² has two advantages over Veitch's method: the reaction between soil and lime occurs in an approximately neutral and un-heated solution, and the work is more easily done. We have as yet insufficient data for judging the importance of the method in determining the lime-requirement of soils, but (as I shall indicate later), we hardly dare take for granted that the ability of the soil to absorb lime determines the degree of its lime-requirement.

In 1909, R. Albert (1) in Greifswalde published the following method for making a quantitative determination of the acidity of the soil.

To soil placed in water add a definite amount of baryta-water and ammonium chloride. Drive off by boiling the ammonium freed by the action of the baryta-water on the ammonium salt, and collect in a retort with diluted sulfuric acid. A previous determination should have been made of the amount of ammonium going into the retort when pure materials are used, and the difference between this amount and the amount of ammonium freed after

² These authors, too, take for granted the quite general impression that the degree of the ability of the soil to absorb lime is at the same time a measure for the degree of its acidity and its lime-requirement.—H. R. C.

soil has been added measures the amount of baryta necessary for neutralizing the soil acids—or expresses the acidity of the soil.

Besides being much simpler and more easily carried out than the Tacke-Süchtung method, this method claims the additional advantage of being able to express the measure of the basicity of the soil, for when basic soils are used, more ammonium is freed than corresponds to the amount of baryta water used.

J. A. Bizzell and T. L. Lyon (5) who also consider the determination of the power of the soil to absorb lime to be the best method for expressing quantitatively the lime-requirement of the soil, have recently attempted to better Albert's method. They heat the mixture of baryta-water and soil for an hour in a boiling water-bath before adding ammonium chloride and making the subsequent distillation. They have proved that the absorption of barium does not take place as quickly as Albert took for granted. Moreover, these scientists prove that soil heated with a pure ammonium chloride solution is able, to a certain extent, to free ammonium from this, but as the ability varies in the various soils, it is necessary in each case to correct the results obtained by distillation with baryta-water with that obtained by the named blind determination.

As Süchtung and Arndt (28) have pointed out, the method used by Albert, and which at first glance appeared so attractive, is wrong in principle; for the strongly alkaline hydroxides (especially on the application of heat) decompose the organic substances in the soil. Therefore, methods based on this principle cannot be used for making an exact determination of the acidity or basicity of the soil.

In the papers appearing in the two years 1909 (2) and 1910 (4), A. Baumann and his collaborator, E. Gully, basing their opinion on results from modern colloidal chemistry as well as on their own wide researches, insist, especially in the case of sphagnum-peat, that it is incorrect to speak of acidity in general, for the acid effects of that substance (for instance its ability to color litmus paper red), should be explained as surface effects (adsorption effects). In sphagnum-peat it is not a question of acidity determination but merely of its power to adsorb bases (or what is the same thing—to free acids).³ A good expression of this power, according to Baumann and Gully, is to treat peat with a calcium acetate solution and determine the amount of acid freed by the inter-action of these substances.

As in the opinion of the above-named scientists, the freeing of acetic acid is solely due to surface effects, this for the most part, quantitatively seen,

³ As I have shown in a previous paper (9) Baumann and Gully are not entirely right. In the electro-metric measurements which the author of this present article in collaboration with J. Witt and N. Feilberg has made of hydrogen-ion concentration in mixtures of raw sphagnum peat and water, a considerable number of hydrogen-ions were found present. G. Fischer (13) attained a similar result from electro-metric determinations of hydrogen-ion concentrations in various samples of sphagnum-peat from German bogs.

will depend on the relative amounts of soil and calcium acetate solution used. An accurate expression for the absolute power to free acids will, therefore, be obtained only by varying these amounts. When, following the experiments of the above-named scientists, we use a limited amount of peat (3 gm.) with a very large amount of concentrated calcium acetate solution,⁴ we obtain an approximately correct expression for the absolute power to free acids; for under these conditions possibilities have been brought about for an approximately maximum adsorption of the base of the salt named, and the results of the acid-determinations carried out in this manner seem to coincide very closely with the results of acid-determinations made with the use of diluted sodium hydroxide.

In mineral soils, the use of solutions of acetates for acidity determinations has been proposed by Oscar Loew (20), who uses the following method.

Fifty grams of air-dry pulverized soil are placed in a 200 cc. neutral 1 per cent solution of sodium or potassium acetate. The mixture should stand (shaken, however from time to time), for 24 hours in a normal room temperature. It is then filtered and of the filtrate 100 cc. measured off for titration.

By the help of this method, Loew claims that an approximate determination of the amount of lime necessary for neutralizing the soil can be made.

Ach. Grégoire (15) and his collaborators have attempted to determine the reaction of the soil by shaking it with a mixture of solutions of potassium-iodate, potassium iodide and sodium hyposulfite, proposed by Kjeldahl in his acidi-metric determinations, and titrating the iodine freed in reaction with free soil acids with a sodium hyposulfite solution, using starch-water as an indicator. By this method the authors have demonstrated that mineral soils may vary greatly in acidity. They mention the fact that silicic acid and zeolites do not affect the reagent, while, on the other hand, this is very sensitive to calcium bicarbonate. Sodium bicarbonate affects it to a very slight degree.

Stutzer and Haupt (24) have applied this principle in the quantitative determination of the reaction of the soil, and give a detailed account of the method for such a determination. In another paper (25), the above-mentioned authors draw attention to the fact that the application of the principle is limited, for iodine may also be freed by ferri-salts, cupri-salts, free chlorine, sodium hypochlorite, and—in the presence of acids—by oxidizing chemical combinations; for instance, peroxides, potassium permanganate and potassium bichromate.

⁴ In a later publication of E. Gully (16), it appears that in the experiments referred to here, as to the ability of sphagnum-peat to free acids by means of the lime-acetate method, 3 grams of air-dry peat has been used with 200 cc. of 10 per cent solution of calcium acetate, and a three-hour reaction time.

In the article in which Baumann and Gully (4) for the first time present the idea of using calcium acetate in determining the ability of sphagnum-peat to free acid, nothing is said as to whether the weight of peat used (3 grams), referred to fresh moist peat, or to dry peat.

Finally we must mention J. G. Lipman's proposal (19). In determining the acidity of the soil he uses a micro-biological method, based on the sensitiveness of certain bacteria to the presence of free acids in the substratum.

A series of equally large portions of a neutral bouillon are measured off. In each place a different quantity of the soil sample under examination (for instance, 0.5 gram, 1 gram, 3 grams, 5 grams, 10 grams). Sterilize all the bouillon portions, and when cool infect with the standard bacterium chosen (*Bacillus mycoides* or *Bacillus subtilis*). After a certain length of time has elapsed, determine the degree of bacteria development in the bouillon culture by measuring the amount of ammonium found. If, for the sake of comparison, similar experiments are made with bouillon portions minus soil, in which the degree of acidity varies, due to the direct addition of varying amounts of acid to each portion, it is possible—within a certain limit—to determine to what amount of acid the development of a definite amount of ammonium by a definite quantity of soil corresponds.

According to Lipman's observations, the above-mentioned species of bacteria can develop in an acid content of up to 2 per cent of the weight of the bouillon. In these experiments it is possible also to use *Azotobacter* with a mannite solution for their nourishment.

Lipman promises further accounts of experiments based on this principle, and not until these appear can we form a positive estimate of the value of this method for determining the acidity of the soil. However, since in addition to the acidity various other soil characteristics exert a great influence on the development of bacteria, and on the decomposition of the nourishment substrata already mentioned [see further Harald R. Christensen (9)], it would appear in advance that this method is not exact enough. Even could all such difficulties be eliminated, the results obtained—especially from the bouillon-culture—seem rather to express the hydrogen-ion concentration in the substratum than the absolute quantity of acid, for it appears to be the latter alone which determines the extent of the enzymatic decompositions [see further S. P. L. Sørensen (23)]. The necessity for sterilizing the bouillon portions after the soil has been added is a weakness in the method, in view of the chemical changes in the soil caused by heat.

Without at this point going further into the discussion of the much disputed question of the existence of humus acids, the author wishes to express as his opinion, that in view of the results obtained from the chemical experimental methods described above, it is more correct to speak of *acid-freeing power* or *base-adsorption power* of the soil, than of the *acidity* of the soil. To determine this last-mentioned power, it will probably be necessary to use other principles, than those in general use at the present time. Possibly experiments with hydrogen-ion concentration will give valuable information in this connection, even though in agricultural soil analyses such determinations could scarcely take the place of determinations of the absolute acid content of the soil.

In the investigations described below only those methods have been used which are able to express—more or less completely—the ability of the soil to free acids from salts. As the method used by Baumann and Gully (4) (the acetate method) seemed to the author to be the simplest and most attractive of the methods hitherto proposed, particular emphasis has been placed upon

a fuller test of the feasibility of adopting this method, and upon giving it such a form that it may be used under the greatly varying conditions in agricultural soil investigations.

2. The use of the calcium acetate method in determining the ability of the soil to free acids

As various soil chemists, M. Fleischer (14), Baumann and Gully (4), and others have shown, the amount of acid which a definite quantity of peat is able to free in a neutral salt solution is determined to a large extent by the relation between the amount of peat and the amount of salt solution used. In regard to the freeing of acetic acid from acetates, Baumann and Gully (4, p. 125) have made absorption experiments using a 25 per cent solution of sodium acetate with fresh sphagnum, as well as with sphagnum-peat, and shown that this ability diminishes perceptibly when the quantity of these last-named substances is increased.

My object is first and foremost to throw light upon the extent to which this relationship appears when the calcium acetate method is used with various soils.

As mentioned before page 121, the absolute value of the ability to free acids is approached the more closely, the larger the difference between the amount of peat and of acetate solution used. However, as the determination (titration) of the acid freed, may be made with the greater accuracy, the larger the amount, it is important not to make the difference too small. Closely connected with these investigations, others have been made as to the influence of the reaction-time on the freeing of acids, and finally as to the influence of air-drying on this ability.

These investigations include both humus soils and ordinary field soils (mineral soils).

The method used is as follows:

Place the soil when weighed off, in a retort of about 500 cc. capacity and cover with exactly 300 cc. of calcium acetate solution (1 gram of calcium acetate to 10 cc. of distilled water). Cork the retort and shake from time to time during the entire reaction-period. When the prescribed length of time has elapsed, filter the solution and titrate (with 1/10 N sodium hydrate) using phenolphthalein as an indicator.

In titration 50 to 100 cc. of the filtrate is used, preferably the former amount, as the change is the more distinct the smaller the amount of fluid. By titrating 3 or 4 portions from the same filtrate, and taking the average of the single titrations, even a very slightly acid content can be estimated with great accuracy. It is important not to use too much phenolphthalein as the most distinct change appears when a relatively small amount (3 to 4 drops of a 1 per cent solution) is used. Phenolphthalein causes precipitation in a calcium acetate solution, and therefore the change of color is not so distinct as when (for instance) sulfuric acid is titrated. Dropping with sodium hydrate must cease at the first suggestion of a change in color (very

slight dirty red tinge). Duplicate determinations were always made (duplicate retorts being used), besides, as was mentioned before, taking 3 to 4 titrations of the filtrates from each retort. As the watery solution of calcium acetate of commerce⁵ is neutral only by exception, and usually shows a slightly acid or alkaline reaction for phenolphthalein, a blind determination of the reaction in the solution used must be taken, and the value found either added to, or subtracted from the values obtained by titrating the filtrates from the mixture⁶ of soil and water.

The power to free acid is, in every investigation made, expressed by the amount of acid—given in the number of cc. 1/10 N acid⁷—which 1 gram of dry soil is able to free in the calcium acetate solution used.

a. Investigations as to the influence which the relation between the amount of soil and of calcium acetate solution, as well as the reaction-time, exert on the ability of the soil to free acid. In these investigations, high-bog peat⁸ and low-bog peat⁹ from the bogs near Studsgaard and Tylstrup, silt-soil from meadows near Varde, and acid field soils have been used.

The moist peat soils were ground in an ordinary meat chopper. This treatment, which presents no difficulty, and a subsequent stirring gave the material a homogenous form and made it easy to handle. Field soils, like the peat soils, were used in their natural moist condition, the various samples thoroughly mixed and possible small pebbles thrown away before being weighed and placed in the retorts. No other special preparation was made.

In table 1, are the results of a series of investigations in which both the amount of soil and the reaction-time were varied simultaneously, while the amount of liquid (300 cc.) remained constant. The results are shown graphically in figure 1.

The investigations were made in two series.

If we turn first to the results of the investigations in Series I, we see that both the quantity of the soil and the reaction-time exert an important influence on the extent to which acid is freed.

Agreeing with the report from Baumann and Gully (4), we too find the largest amount of acid free per weight unit of soil when the smallest amount of peat soil is used. But as the table shows, by prolonging the reaction-time we possess a means for eliminating totally or partially those differences which appear when varying quantities of soil are used. Judging by the results obtained from peat soils, a five-hour reaction-time seems to yield approximately the maximum amount of acid freed, if the amount of peat used is not larger than a corresponding 0.5 gram to 0.75 gram of dry soil. When larger quantities of peat are used the values for the amount of acid freed, gen-

⁵ The calcium-acetate used in the investigations comes from Kahlbaum, Berlin.

⁶ In Baumann and Gully's investigations the solution is neutralized before being used.

⁷ Danish—Hojmosetrø; German—Hochmoortorf.

⁸ Danish—Lavmosetrø; German—Niedermoortorf; English—fen (according to Edw. J. Russell, Soil Conditions and Plant Growth, p. 69–70).

erally seem to diminish appreciably. For field (mineral) soils, a five-hour reaction-time seems to yield approximately the maximum amount of acid freed per weight unit of soil, when an amount up to 10 to 15 grams of dry soil is used. Were the reaction-time but three hours, only 5 grams of dry soil could be used when proportionately just as large an amount of acid is to be freed.

Later, certain observations of the influence of the reaction-time on the amount of acid freed seemed to indicate that a five-hour reaction-time was not sufficient for all soils to bring the reaction between the soil and the calcium acetate solution to a close. New investigations (table 1, series 2; and

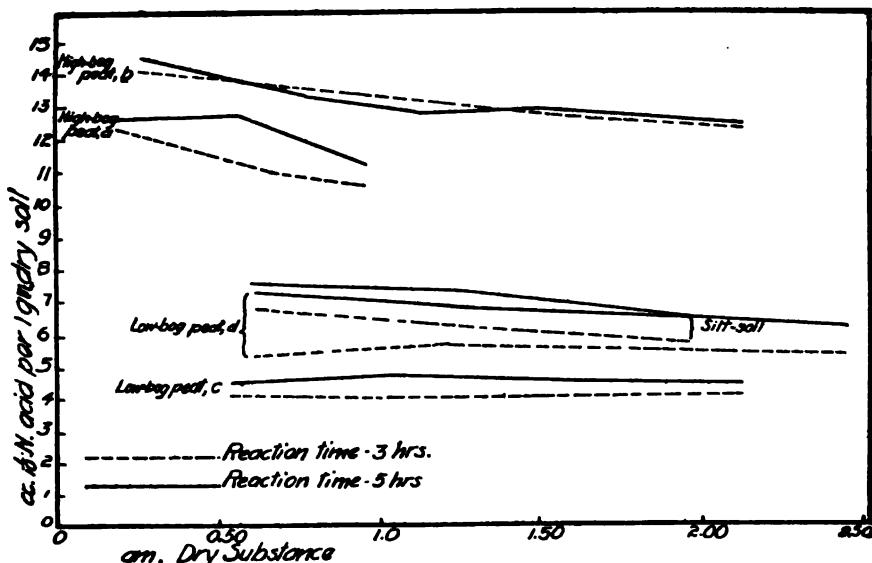


FIG. 1. DIAGRAM SHOWING THE INFLUENCE OF THE AMOUNT OF SOIL AND THE REACTION-TIME ON THE AMOUNT OF ACID FREED FROM CALCIUM ACETATE SOLUTION

table 2) were made, high-bog peat, low-bog peat, mineral soils and silt soil being used.

The results of these investigations establish the fact that the reaction-time as well as the proportional amounts of soil and salt solution used, have a great influence upon the degree of the amount of acid freed, and as may be seen from the tables, of these two factors, the reaction-time is usually the more important.⁹ In the case of high-bog peat this factor is of relatively

⁹ Though in the case of colloid material it is difficult to determine to what extent we are dealing with adsorption or with chemical reaction, it is generally safe to say that the first process ceases quite quickly. When such a heterogeneous material as soil is used, of which the larger part consists of combinations very difficult of solution, a very long time may elapse before the chemical equilibrium is established. The great influence which the reaction-time in these experiments exerts on the absorption of bases may perhaps indicate that this absorption is not occasioned by surface effects alone.

TABLE 1
The influence of the amount of soil used and the reaction time on the ability of the soil to free acids
Series I

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry ^a soil
Fresh, moist	Dry ^a		The single retorts†	Average		
High bog peat A from "Knudemose" near Herning (strong acid reaction)						
3	0.30	3	1.26 1.23	1.25	1.25	12.5
6	0.60	3	2.26 2.31	2.29	1.15	11.5
9	0.90	3	3.21 3.31	3.26	1.09	10.9
3	0.30	5	1.28 1.26	1.27	1.27	12.7
6	0.60	5	2.55 2.47	2.51	1.27	12.7
9	0.90	5	3.50 3.45	3.48	1.16	11.6
High-bog peat B from "Store Vildmose" near Tylstrup (strong acid reaction)						
3	0.36	3	1.67 1.67	1.67	1.67	13.9
6	0.72	3	3.26 3.26	3.26	1.63	13.6
9	1.08	3	4.64 4.74	4.69	1.56	13.0
12	1.44	3	6.09 6.14	6.12	1.53	12.7
18	2.16	3	8.85 8.80	8.83	1.47	12.3
3	0.36	5	1.75 1.70	1.73	1.73	14.4
6	0.72	5	3.24 3.21	3.23	1.62	13.5

TABLE 1 (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETRES 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry* soil
Fresh, moist	Dry*		The single retorts†	Average		
High-bog peat B from "Store Vildmose" near Tylstrup (strong acid reaction)						
9	1.08	5	4.63 4.61	4.62	1.54	12.8
12	1.44	5	6.17 6.14	6.16	1.54	12.8
18	2.16	5	8.91 8.99	8.95	1.49	12.4
Low-bog peat A from "Store Vildmose" (strong acid reaction)						
1½	0.26	3	0.71 0.71	0.71	1.42	8.2
3	0.52	3	1.41 1.44	1.43	1.43	8.3
4½	0.78	3	2.13 2.13	2.13	1.40	8.1
6	1.04	3	2.94 2.85	2.90	1.45	8.4
1½	0.26	5	0.71 0.71	0.71	1.42	8.2
3	0.52	5	1.31 1.41	1.36	1.36	7.9
4½	0.78	5	2.09 2.07	2.08	1.39	8.0
6	1.04	5	2.91 2.84	2.87	1.44	8.3
Low-bog peat B from "Tylstrup" (strong acid reaction)						
3	0.57	3	1.70 1.65	1.68	1.68	8.8
6	1.14	3	3.23 3.26	3.25	1.63	8.6

TABLE 1 (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry ^a soil
Fresh, moist	Dry ^a		The single retorts†	Average		
Low-bog peat B from "Tylstrup" (strong acid reaction)						
9	1.71	3	4.36 4.36	4.36	1.45	7.6
12	2.28	3	5.62 5.67	5.65	1.41	7.4
18	3.48	3	7.69 7.82	7.76	1.29	6.8
24	4.56	3	9.72	9.72	1.22	6.4
Good sandy soil (acid reaction)						
3	2.6	3	0.65 0.65	0.65	0.65	0.75
6	5.2	3	1.21 1.21	1.21	0.61	0.70
9	7.8	3	1.80 1.90	1.85	0.62	0.72
12	10.4	3	2.22 2.17	2.20	0.55	0.63
15	13.0	3	2.41 2.36	2.39	0.48	0.55
15	13.0	3	2.71 2.71	2.71	0.54	0.63
15	13.0	5	2.86 2.81	2.84	0.57	0.66
Clay soil (A) (acid reaction)						
3	2.5	3	0.55 0.60	0.58	0.58	0.70
6	5.0	3	0.98 1.03	1.01	0.51	0.60

TABLE I (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid	The single retort†	Average	Per 1 gm. moist soil
Fresh, moist	Dry*					
Clay soil (A) (acid reaction)						
9	7.5	3	1.26 1.40	1.33	0.44	0.53
12	10.0	3	1.70 1.85	1.78	0.45	0.53
18	15.0	3	2.34	2.34	0.39	0.47
3	2.5	5	0.53 0.53	0.53	0.53	0.64
6	5.0	5	1.00 1.02	1.01	0.51	0.60
9	7.5	5	1.56 1.51	1.54	0.51	0.62
12	10.0	5	2.11 2.05	2.08	0.52	0.62
18	15.0	5	2.91 2.91	2.91	0.49	0.58
Series 2						
High-bog peat C from "Knunderose" (strong acid reaction)						
6	0.88	3	2.95 3.11	3.03	1.52	10.3
3	0.44	5	1.73 1.69	1.71	1.71	11.6
6	0.88	5	3.21 3.19	3.20	1.60	10.9
12	1.76	5	6.06 5.86	5.96	1.49	10.2
Low-bog peat C from Gelleruplund (acid reaction); (6/10 1913)						
2	0.542	3	0.73 0.70	0.72	1.08	4.0

TABLE 1 (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry* soil
Fresh, moist	Dry*		The single retorts†	Average		
Low-bog peat C from Gelleruplund (acid reaction) (6/10 1913)						
4	1.084	3	1.44 1.40	1.42	1.07	3.9
6	1.626	3	2.15 2.19	2.17	1.09	4.0
8	2.168	3	2.90 2.90	2.90	1.09	4.0
12	3.252	3	4.26 4.24	4.25	1.06	3.9
2	0.542	5	0.77 0.81	0.79	1.19	4.4
4	1.084	5	1.72 1.78	1.75	1.31	4.9
6	1.626	5	2.52 2.54	2.53	1.27	4.7
8	2.168	5	3.41 3.41	3.41	1.28	4.7
12	3.252	5	4.93 4.91	4.92	1.23	4.5
Same low-bog peat (8/10 1913)						
2	0.542	3	0.72 0.72	0.72	1.08	4.0
4	1.084	3	1.56 1.64	1.60	1.20	4.4
8	2.168	3	3.04 3.12	3.08	1.16	4.2
2	0.542	5	0.85 0.84	0.85	1.28	4.7
4	1.084	5	1.76 1.72	1.74	1.31	4.8

TABLE 1 (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry ^a soil
Fresh, moist	Dry ^a		The single retorts†	Average		
Same low-bog peat (8/10 1913)						
8	2.168	5	3.45 3.41	3.43	1.29	4.7
12	3.252	5	5.08 5.02	5.05	1.29	4.7
2	0.542	7	0.95 0.91	0.93	1.40	5.1
4	1.084	7	1.97 1.89	1.93	1.45	5.3
8	2.168	7	3.72 3.82	3.77	1.41	5.2
Low-bog peat D from "Store Vildmose" (strong acid reaction)						
2.1	0.612	3‡	1.15		1.64	5.6
4.2	1.224	3	2.35		1.68	5.8
8.4	2.448	3	4.40 4.48	4.44	1.59	5.5
16.8	4.896	3	8.59 8.41	8.50	1.52	5.2
2.1	0.612	5	1.47		2.1	7.2
4.2	1.224	3	2.68 2.82	2.75	1.96	6.7
8.4	2.448	3	5.15 5.15	5.15	1.84	6.3
16.8	4.896	3	9.21 9.19	9.20	1.64	5.6

TABLE 1 (Cont.)

GRAMS SOIL USED		REACTION TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID			
			Per 100 cc. liquid		Per 1 gm. moist soil	Per 1 gm. dry ^a soil
Fresh, moist	Dry ^a		The single retorts ^b	Average		
Silt soil no. 12 (acid reaction)						
3	0.62	3	1.39 1.43	1.41	1.41	6.8
6	1.24	3	2.57 2.63	2.60	1.30	6.3
9	1.86	3	3.67 3.67	3.67	1.22	5.9
3	0.62	5	1.58 1.54	1.56	1.56	7.5
6	1.24	5	2.90 2.92	2.91	1.46	7.1
9	1.86	5	3.98 4.02	4.00	1.33	6.5
Loam soil, poor in humus (B) (strong acid reaction)						
5	4.6	3	0.66 0.72	0.69	0.41	0.45
10	9.2	3	1.23 1.25	1.24	0.37	0.40
20	18.4	3	2.09 2.09	2.09	0.31	0.34
10	9.2	5	1.29 1.29	1.29	0.39	0.42
5	4.6	7	0.78 0.76	0.77	0.46	0.50
10	9.2	7	1.34 1.30	1.32	0.40	0.43
20	18.4	7	2.29 2.29	2.29	0.34	0.37

* Here, and in the following investigations, "dry" applied to peat soils means "oven-dry," applied to field soils, "air-dry."

† Here and later, after correction has been made for content of water in the soil.

‡ Due to an oversight the reaction-time was 20 minutes too long.

small importance, even though when larger quantities of peat are used, there is a distinct difference between a three and a five-hour reaction-time; in high-bog peat, specimens A and C, increasing the reaction-time has annulled the difference in amounts of acid freed, caused by using various quantities of soil. In high-bog peat, specimen B, the amount of acid freed is just as large after a three-hour as after a five-hour reaction-time, and this experiment merely expresses the variations caused by the differences in the amount of peat used. However, these variations are comparatively small, not only here but also in other specimens of high-bog peat examined.

Low-bog peat C is a clear example of a type of soil in which the relation between the amounts of soil and of calcium acetate solution has no influence on the amount of acid freed, which, on the other hand, is largely determined by the length of the reaction-time. As appears in table 1 (series 2) the various amounts of peat (from 0.54 gram to 3.25 grams of dry peat soil) used have freed an equally large amount of acid per weight unit during the various reaction-times. The same experiment also proves that even a reaction-time of five hours was insufficient to complete the base-absorption, for during an additional two hours a very appreciable increase in the amount of acid freed was observed. (A further discussion of this will follow later.)

In low-bog peat D it was apparent that in a three-hour reaction-time approximately the same amount of acid was freed per weight unit of peat, even though the amount of peat used varied between 0.61 gram to 4.9 grams of dry soil. If the reaction-time was increased to five hours in all the samples of peat examined, the amount of acid freed was likewise found to increase, but the amount varied greatly in the various samples. This specimen of low-bog peat acts otherwise than specimen A from the same bog and which freed the same amount of acid even when the reaction-time was increased (over three hours), and the amount of peat varied.

In the case of silt soil no. 12, and the two loam soils, both the amount of soil used and the reaction-time have influenced the extent of acid freed.

In loam soil A an increase in the reaction-time from three to five hours was sufficient to almost entirely eliminate the difference in amount of acid freed caused by using various amounts of soil. This was not true in loam soil B, for after a period of seven hours the difference was still very apparent. On account of the limited power of this soil to free acid, the differences in amount of acid freed during the reaction-times tested and with the smaller quantities of soil used are so small that the titration method in use is powerless to determine them exactly. Yet the absolutely as well as relatively appreciable difference in acid freed per weight unit of soil between the largest amount of soil (20 grams), and the smallest amount (5 grams) used, indicates positively that in this type of soil, in contrast with loam soil A, the amount of acid freed is more largely determined by the amount of soil used than by the length of the reaction-time.

In order to illustrate further the extent to which the freeing of acid is de-

terminated by the reaction-time, the experiments referred to in table 2 were made, and using the same quantity of soil the reaction-time was varied within very wide limits. In some of the experiments the same soil was used as in the experiment just referred to (table 1).

If in series 1, table 2, we examine the relation between humus soils and their reaction time, we see that in high-bog peat acid is freed much more quickly than in low-bog peat. When one-eighth of an hour has passed the acid freed from high-bog peat has attained 70 per cent of the total amount of acid freed during the ten-hour reaction-time; whereas low-bog peat from Tylstrup and low-bog peat from Gelleruplund yield in the same time 32 and 29 per cent, respectively.

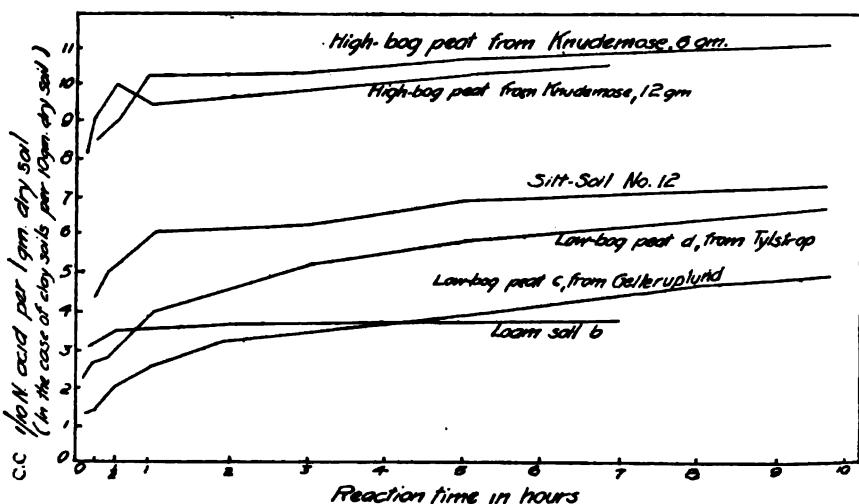


FIG. 2. DIAGRAM SHOWING THE INFLUENCE OF THE REACTION-TIME ON THE ABILITY OF VARIOUS SOILS TO FREE ACID FROM A CALCIUM ACETATE SOLUTION

Base-absorption by low-bog peat is particularly slow and is only approximately brought to an end after eight hours. In high-bog peat, silt soil and loam soil the reaction is approximately ended after five hours have passed.

It is especially interesting to note that base-absorption by high-bog peat B is quite independent of the quantity of peat used, for relatively speaking, almost the same amount is absorbed whether 12 grams of peat are used or 6 grams. The specimen of high-bog peat under discussion, belongs, therefore, to that type of soil in which the relation between the amount of soil and the salt solution is of subordinate importance in the amount of acid freed, and differs from high-bog specimen A previously examined and coming from the same bog (see table 1). In the latter case the amount of peat used exerted not a great, but yet a very distinct influence on the degree of acid freed.

In figure 2 is a diagram showing the relation between the reaction-time and the base-absorption in the various soils examined.

TABLE 2

*Influence of the reaction-time on the ability of the soil to free acid, the amount of soil used being constant
Series 1*

REACTION-TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID					
	Per 100 cc. liquid			Per 1 gram moist soil	Per 1 gram dry soil	
	Retort a	Retort b	Average			
High-bog peat B from "Knudemose" (strong acid reaction); 6 grams fresh, moist peat soil (0.88 gram dry soil) used in each retort						
½	2.29	2.22	2.26	1.13	7.71	
¼	2.64	2.41	2.53	1.27	8.63	
½	2.53	2.66	2.65	1.33	9.04	
1	2.93	3.03	2.98	1.49	10.17	
2	3.09	2.64	(2.92)*	(1.46)*	(9.97)*	
3	2.95	3.11	3.03	1.52	10.34	
5	3.21	3.19	3.20	1.60	10.92	
7	3.27	3.18	3.23	1.62	11.02	
10	3.25	3.29	3.27	1.64	11.16	
Same high-bog peat; 12 grams fresh moist peat soil (1.76 grams dry soil) used in each retort						
½	4.72	4.89	4.81	1.20	8.19	
¼	5.18	(3.43)†		1.30	8.82	
½	5.97	5.68	5.83	1.46	9.93	
1	5.70	5.30	5.50	1.38	9.37	
2	5.89	5.49	5.69	1.42	9.69	
3	5.91	5.79	5.85	1.46	9.97	
5	6.20	6.26	6.23	1.56	10.61	
7	6.41	6.35	6.38	1.60	10.87	
Low-bog peat D from Tylstrup (strong acid reaction); 8.14 grams fresh, moist peat (2.447 grams dry soil) used in each retort						
½	1.85			0.66	2.27	
¼	2.28	2.26	2.27	0.81	2.78	
½	2.38	2.34	2.36	0.84	2.89	
1	3.41	3.20	3.31	1.18	4.06	
3	4.39	4.47	4.43	1.58	5.43	
5	5.15	5.15	5.15	1.82	6.31	
7	5.40	5.48	5.44	1.94	6.66	
10	5.68	5.74	5.71	2.04	7.00	
Low-bog peat C from Gelleruplund (acid reaction); 9 grams fresh, moist peat (2.439 grams dry soil) used in each retort						
½	1.32	1.15	1.24	0.41	1.53	
¼	1.34	1.40	1.37	0.46	1.69	
½	1.79	1.85	1.82	0.61	2.24	
1	2.33	2.27	2.30	0.77	2.83	

TABLE 2 (Cont.)

REACTION-TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID				
	Per 100 cc. liquid			Per 1 gram moist soil	Per 1 gram dry soil
	Retort a	Retort b	Average		
Low-bog peat C from Gelleruplund (acid reaction) 9 grams fresh, moist peat (2.439 grams dry soil) used in each retort					
2	2.76	2.82	2.79	0.93	3.43
4	3.41	3.42	3.42	1.14	4.19
6	3.75	3.75	3.75	1.25	4.61
8	4.10	4.06	4.08	1.36	5.02
10	4.20	4.18	4.19	1.40	5.15
11	4.28			1.43	5.26
Silt soil no. 12 (acid reaction); 6 grams fresh moist soil (1.24 gram dry soil) used in each retort					
½	1.92	1.78	1.85	0.93	4.48
½	2.02	2.30	2.16	1.08	5.21
1	2.41	2.45	2.43	1.22	5.88
3	2.57	2.63	2.60	1.30	6.30
5	2.90	2.92	2.91	1.46	7.05
7	2.94	2.94	2.94	1.47	7.12
10	2.98	3.08	3.03	1.52	7.34
Loam soil B (strong acid reaction); 10 grams fresh moist soil (=9.2 grams air-dry soil) used in each retort					
½	0.94	0.96	0.95		0.31
½	0.98	1.06	1.02		0.33
½	1.10	1.14	1.12		0.37
1	1.18	1.16	1.17		0.38
2	1.20	1.26	1.23		0.40
3	1.24	1.26	1.25		0.41
5	1.30	1.30	1.30		0.42
7	1.36	1.32	1.34		0.44
High-bog peat from "Knudemose" (strong acid reaction); 7.6 grams fresh moist peat (=1 gram dry soil) used in each retort					
Shaken from time to time (usual method of procedure)					
3	3.35	3.29	3.32		9.96
5	3.37	3.39	3.38		10.14
10	3.43	3.45	3.44		10.32
20	3.48	3.52	3.50		10.50

TABLE 2 (Cont.)

REACTION-TIME IN HOURS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID					
	Per 100 cc. liquid			Per 1 gram moist soil		
	Retort a	Retort b	Average			
High-bog peat from "Knudemose" (strong acid reaction) 7.6 grams fresh moist peat (=1 gram dry soil) used in each retort						
Unshaken						
3	3.28	3.27	3.28		9.84	
5	3.33	3.37	3.35		10.05	
10	3.41	3.39	3.40		10.20	
20	3.52	3.46	3.49		10.47	
Low-bog peat from Gelleruplund (reaction undetermined); 6 grams fresh moist peat (=1.5 gram dry soil) used in each retort						
Shaken from time to time						
3	2.31	2.37	2.34		4.68	
5	2.57	2.59	2.58		5.16	
10	3.12	3.12	3.12		6.24	
20	3.18	3.06	3.12		6.24	
19†	3.10	3.16	3.13		6.26	
Unshaken						
3	2.22	2.12	2.17		4.34	
5	2.47	2.45	2.46		4.92	
10	2.80	2.88	2.84		5.68	
20§	3.06	3.08	3.07		6.14	
19†	3.00	2.96	2.98		5.96	
Clay soil, poor in humus (acid reaction); 11.55 grams fresh moist soil (= 10 grams air-dry soil) used in each retort						
Shaken from time to time						
3	1.27	1.31	1.29		0.387	
5	1.39	1.37	1.38		0.414	
10	1.55	1.51	1.53		0.459	
19	1.51	1.53	1.52		0.456	
Unshaken						
3	1.14	1.04	1.09		0.327	
5	1.20	1.20	1.20		0.360	
10	1.27	1.27	1.27		0.381	
19	1.47	1.47	1.47		0.441	

* On account of a very considerable difference in the results of the duplicate determinations, presumably due to a flagrant error, the correctness of the average is to be disputed.

† The cause of this low—and presumably incorrect—figure is unknown. No account has, therefore, been made of this figure in computing the results per weight measure.

‡ Set aside a few days later.

§ After standing quietly for about 19 hours, the retorts were inadvertently shaken a single time.

In the last section of table 2 are the results of an examination of three different kinds of soils (high-bog peat, low-bog peat, and loam soil), in which the reaction-time varies from three to twenty hours. At the same time the importance of shaking the retorts during the reaction-time has been tested. In the first part of the experiment the retorts were shaken hard quite often¹⁰—about every fifteen minutes; in the second part of the experiment the soil and the liquid were thoroughly shaken together immediately after being placed in the retorts, and again just before being filtered; in the interim the retorts stood quietly on a shelf.

In agreement with the tests referred to in the first section of table 1, we find that the reaction between soil and calcium acetate solution ceases after a

TABLE 3

Investigations of the influence exerted by various amounts of calcium acetate solution on the ability of a certain amount of soil to free acid (reaction-time five hours)

AMOUNT OF SOIL; GRAMS OF DRY SUBSTANCE	AMOUNT OF CALCIUM ACE- TATE SOLUTION IN CUBIC CENTIMETERS	ACID FREED EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID				
		Per 100 cc. liquid			Per 1 gram of dry soil	
		Retort a	Retort b	Average		
<i>High-bog peat</i>						
1	200	4.99	4.95	4.97		9.94
1	300	3.37	3.39	3.38		10.14
1	400	2.58	2.58	2.58		10.32
<i>Low-bog peat</i>						
1½	200	3.73	3.67	3.70		4.93
1½	300	2.57	2.59	2.58		5.16
1½	400	2.03	1.99	2.01		5.36
1½	500	1.64				5.47
<i>Loam soil</i>						
10	200	1.95	1.91	1.93		0.386
10	300	1.39	1.37	1.38		0.414
10	400	1.12	1.12	1.12		0.448

period of about ten hours. Shaking the retorts has had no especial influence on high-bog peat. In the case of low-bog peat, this action has caused an appreciable increase in the amount of acid freed in the reaction-times three, five and ten hours, but in the longest reaction-time (twenty hours), the difference has almost entirely disappeared.

Table 3 shows the results of an absorption-experiment in which the amount of soil is kept constant, while the amount of calcium-acetate solution is varied. In this test the same soils are used as in the one last mentioned.

¹⁰ In the case of the longest reaction-time the retorts were shaken only during the first four to five hours and the last four to five hours before filtering. In the intervening interval they remained undisturbed.

We find that the greatest amount of acid is freed per weight unit of soil when the greatest amount of solution is used. The results agree, as was also to be expected, with those obtained from the tests previously mentioned, in which quantities of soil of varying weight were used with the same amount of calcium-acetate solution.

In order to judge the results, we will express the amount of acid freed in relation to that value found when 400 cc. of the solution is used. Expressed thus, the difference between the acid freed in 200 cc. of the solution and in 400 cc. of the solution is, for high-bog peat 3.7 per cent, for low-bog peat 8 per cent, and for loam soil 13.8 per cent. The difference in amount of acid freed when 300 cc. and 400 cc. are used is 1.7 per cent, 3.7 per cent and 7.6 per cent, respectively.

The influence of the amount of the solution used is greatest in the case of the loam soil. There appears here to be a considerable difference in the amount

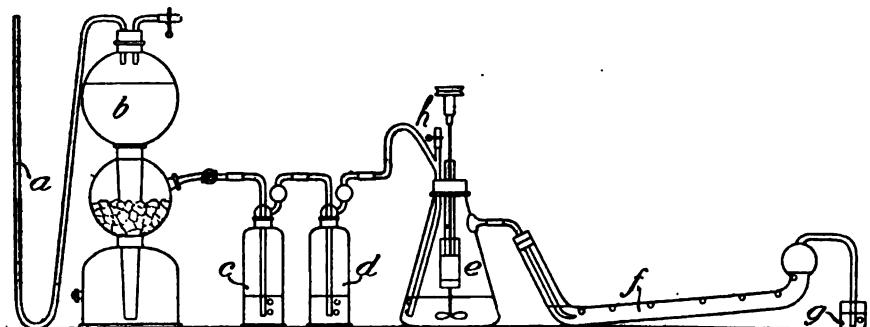


FIG. 3. APPARATUS FOR DETERMINING THE ACIDITY OF THE SOIL ACCORDING TO THE TACKESÜCHTING METHOD

of acid freed according to whether 300 cc. or 400 cc. of the solution is used. In the case of both the humus soils, approximately the maximum amount of acid is freed in the given reaction-time when 300 cc. of the solution is used. A test has been made in which 500 cc. of solution was used with the last-named peat, but this increase in the amount of the solution gives no appreciable change in the acid freed.

As mentioned before, Gully (16), in his latest work in investigating the ability of sphagnum-peat to free acid, has suggested using 3 grams of air-dry peat with 250 cc. of 10 per cent calcium acetate solution in a reaction-time of three hours. When the relation between the amount of peat and the amount of solution is so close, this reaction-time is sometimes insufficient, especially when an approximately absolute expression of the ability of high-bog peat to free acid in calcium acetate solution is to be obtained. According to the investigations referred to in table 2, in which the relation between the amount of peat and the amount of solution has been far wider than Gully suggested, base-absorption from high-bog peat is only approximately com-

plete after five hours have elapsed. Judging further from the results shown in table 1, it is not usually advisable to use more of this nor of any other peat soil, than corresponds to 0.5 to 0.75 gram of dry peat with 300 cc. of solution. Investigations show that for low-bog peat and silt-soil a three-hour reaction-time is quite insufficient. Finally, in order to express in exact figures the ability of mineral-soils to free acid, we must use much larger quantities of soil. When the reaction-time is increased to at least five hours we could, judging by the results of the experiments at hand, advantageously use about 10 grams of soil to 300 cc. of solution.

A wholly satisfactory expression of the power of the soil to free acid from salts can, however, be obtained only by varying the relation between the amount of soil and the amount of solution. This method seems especially advisable when we are dealing with mineral soils. Reference, too, should be made to the calcium-acetate method for determining the ability of the soil to free acid tested by investigations, and following later (p. 149).

b. Investigations of the variations in the ability of soils to free acid from calcium acetate, and the influence which air-drying exerts on this ability. Taking for granted that the ability of the soil to free acid from acetates, is chiefly determined by surface effects, there might be reason to suppose that drying the soil (due to the presence of non-reversible colloids), might cause a diminution of the soil-surface, and therefore of the power of the soil to adsorb bases.

To throw further light on the question the investigations referred to in tables 4 to 6 were made. They include both humus and field (mineral) soils.

All the soil-specimens were examined both in their original moist condition, and after being rendered air-dry. Duplicate determinations were generally made, the method being as follows. The soil was simultaneously weighed off into four retorts. Two of these were immediately filled with 300 cc. of calcium-acetate solution, while the two remaining were first used when the soil in them had been in an air-dry condition during a longer or shorter period (varying from a few days to several weeks).

At the time at which these tests were made, the investigations of the reaction-time mentioned above had been brought only to a temporary conclusion. As these investigations, as we have said before, seemed to indicate that a five-hour reaction-time (a reasonable quantity of soil being used) was sufficient for attaining the approximately maximum amount of acid freed, this reaction-time was chosen. Later investigations showed that this is insufficient for some humus soils, and therefore the results in table 4, are not in every instance absolute. However, they can doubtless express approximately the influence of air-drying on the ability of the soil to free acid from acetates and the variations in that ability.

If we look first at section 1, table 4, which contains results of tests made with raw peat soils, we see that here there is a comparatively small difference

between the ability of peat soil to free acid from calcium acetate before and after air-drying. Yet the results contain the suggestion that high-bog peat and low-bog peat respond differently to air-drying, for two of the three high-bog peat samples examined showed a little smaller, and both of the low-bog peat samples examined a greater ability to free acid when air-dry than when in their original fresh moist condition.

Results from the second section of the table show likewise that air-drying has in the case of low-bog peat caused an appreciable increase in the amount of acid freed. However, the three humus soils, marked "silt soils (nos. 12, 4 and 5), show a different result, for air-drying has to a great extent diminished their power to free acid.

As silt soils, in contrast to peat soils, dry into very hard lumps, it seemed possible that some of the soil in these lumps was never, or in any case, for an insufficient length of time, subjected to the influence of the calcium acetate solution. Therefore, tests were made, referred to in table 5, in pulverizing the soil in a mortar before placing it in the retort.

As the table shows, this precaution has entirely changed the relation. Now the air-dry samples show themselves capable of freeing considerably more acid than the fresh moist samples, and in all three instances the difference is far greater than in the case of any of the low-bog peat samples cited in table 4.

The variation in the ability of humus soils to free acids, is, as table 4 indicates, very large (from 2.2 to 12.7 cc. of 1/10 N acid per gram of air-dry soil). This variation is not only due to a varying content of mineral-soil, but also to a difference in the quality of the humus materials themselves. Low-bog peat generally possesses a much smaller ability to free acid than high-bog peat—due perhaps to the fact that the former humus form is much richer in bases (especially lime) than the latter.

As has been described in greater detail in a previous paper (11), the upper layer of peat (to a depth of 30 cm.), in high-bog peats possesses a far greater ability to free acids than the layer of peat lying just below it (30 to 50 cm.). This difference could not be explained by a difference in base-content, for the lower layer of peat was poorer in lime than the upper. As these peat specimens were in every instance pure peat, with approximately equally large surface (hygroscopicity, determined according to Mitscherlich and Rodewald), it is reasonable to infer that the ability of high-bog peat to free acid is not dependent on surface effects alone, and that the difference in this ability in the two layers of peat is due to a varying content of true acids. [See further article (11), p. 632.]

For the results of the tests made with field soils (mineral soils), see table 6.

As appears from the last column in this table, air-drying has in no single instance diminished the ability of the soil to free acid, but has in each case increased that ability to a distinct degree. In certain instances, especially in the case of soils with small power to free acid, this increase, relatively seen,

TABLE 4

Investigations of the ability of various humus soils to free acids both when fresh and moist and when air-dry (reaction-time five hours)

MARK	GENERAL CONDITION OF THE SOIL	ACID EFFERVESCENCE	REACTION	AZOTOBACTER VEGETATION	GRAMS SOIL USED	ACID FREEED, EXPRESSED IN CUBIC CENTI- METERS 1/10 N ACID USING						DIFFERENCE BETWEEN MOIST AND AIR-DRY PEAT SOIL	
						Fresh, moist peat soil			Air-dry peat soil				
						Per 100 cc. liquid	Per 1 gram dry peat soil	Single reports	Per 100 cc. liquid	Per 1 gram dry peat soil	Single reports		

Series 1. Raw Humus Soils

A	High-bog peat from "St. Vildmose"	None	Strong acid	0	3	0.389	1.76	1.77	13.6	1.64	1.65	12.7	+0.9
II	High-bog peat from "Vejen Mose"	—	—	0	3	0.487	1.46	1.46	9.0	1.42	1.41	8.6	+0.4
M. D. L.	High-bog peat, quite unhumified sphagnum peat	—	—	0	3	0.333	0.90	0.90	8.1	0.94	0.94	8.5	-0.4
B	Low-bog peat from Tylstrup	—	—	0	3	0.952	2.16	2.15	6.8	2.30	2.31	7.3	-0.5
C	Low-bog peat from Gelleruplund	—	?	?	3	0.687	1.20	1.20	5.2	1.30	1.24	5.4	-0.2

Series 2. Cultivated Humus Soils

12	Peat mixed with silt and ochre	None	Acid	0	5	1.02	2.07	2.08	6.12	1.60	1.73	5.09	+1.03
2	Peat soil mixed with sand	—	Slightly acid	0	3	1.18	2.25		5.72	2.29		5.82	-0.10
22	Brownish black peat soil	—	Neutral	1	3	0.97	1.56		4.83	1.62		5.02	-0.19
4	Silt soil	—	Neutral—slightly acid	0	5	1.55	2.22		4.29	1.53		2.96	+1.33
1	Black peat soil mixed with sand	—	Neutral	2	5	1.86	2.36		3.82	2.63		4.25	-0.43
5	Silt soil, not entirely humified	—	Neutral—slightly acid	0	6	2.10	2.46		3.51	1.93		2.76	+0.75

TABLE 4 (Cont.)

MARK	GENERAL CONDITION OF THE SOIL	ACID DIFFUSION	REACTION	AZOTOBACTER VEGETATION	GRAMS SOIL USED	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID USING						DIFFERENCE BETWEEN MOIST AND AIR-DRY PEAT SOIL	
						Fresh, moist peat soil			Air-dry peat soil				
						Per 100 cc. liquid	Per 100 cc. dry	Single retorts	Per 100 cc. liquid	Per 100 cc. dry	Average		
Fresh, moist													
Dry soil													

Series 2. Cultivated Humus Soils

15	Black peat soil mixed with sand, not entirely humified	—	Neutral—slightly alkaline	2	5	1.65	1.83		3.33	1.81		3.30	+0.03
6	Black peat soil mixed with sand	—	Neutral	4	6	2.50	1.63		1.96	1.87		2.24	-0.28
21	Black peat soil mixed with sand	—	Neutral	4	5	1.82	1.30		2.13	1.40		2.30	-0.17

TABLE 5

Investigations of the influence of air-drying and pulverization on the ability of silt soils to free acids (reaction-time five hours)

MARK	Grams soil used	SERIES 1						SERIES 2						DIFFERENCE BETWEEN FRESH, MOIST AND AIR-DRY PULVERIZED SOIL		
		Acid freed, expressed in cubic centimeters 1/10 N acid using:						Acid freed, expressed in cubic centimeters 1/10 N acid, using air-dry and pulverized soil								
		Fresh moist soil			Air-dry (lumpy) soil			Grams soil used			Per 100 cc. liquid					
		Per 100 cc. liquid	The single retorts	Average	Per 100 cc. liquid	The single retorts	Average	Per 100 cc. liquid	The single retorts	Average	Per 100 cc. liquid	The single retorts	Average			
12	5	0.877	1.86	1.86	6.37	1.32	1.31	4.49	+1.88	0.97	0.837	2.10	2.12	7.60	-3.11	-1.23
			1.86			1.30						2.14				
4	5	1.243	1.82	1.84	4.44	1.32	1.24	3.00	+1.44	1.37	1.252	2.08	2.06	4.94	-1.94	-0.50
			1.86			1.16						2.04				
5	5	1.575	2.02	2.00	3.81	1.54	1.55	2.95	+0.86	1.73	1.586	2.42	2.42	4.57	-1.62	-0.76
			1.98			1.56						2.42				

is very considerable. The last two soils mentioned in the table, nos. 1262 and 136, show that the power to free acid is, respectively, two and four times as great in air-dry as in moist soil. Air-drying has just the opposite effect on the majority of humus and mineral soils from what we expected to find (see introduction, p. 140).

It is very interesting to note that all the soils examined, even those effervesing strongly for acid and showing a strong alkaline reaction for litmus, possess a base-absorbing power.

In his "Bodenkunde," published in 1911, E. Ramann (21, p. 242), distinguishes between absorptively un-saturate ("absorptiv ungesättigte"), and absorptively saturate ("absorptiv gesättigte"), soils. In the first group he places the soils hitherto called "acid," and he defines them as "soils which are capable of coloring blue litmus paper red, and freeing acids in solutions of neutral salts." In the second group he places those soils which are base-saturate, and therefore incapable of absorbing bases. In the first group Ramann distinguishes further between neutrally reacting soils, which are only slightly stable, and will sooner or later become absorptively unsaturated, and alkaline reacting soils (those containing carbonate of lime or ammonium carbonate).

As the results of the investigations previously mentioned show, the premises for this division of the soils are incorrect, for we find that not only soil with an acid reaction for litmus, but also those with a neutral or alkaline reaction, possess the ability to absorb bases, and in an ordinary litmus test the means for determining whether or not the soil is "absorptively saturate," are wanting.

The fact that there is no close connection between the ability of the soil to free acid from calcium-acetate and its relation to litmus solution, and that soil may have a very great ability to free acid and still show no acid reaction for litmus,¹¹ seems to suggest that the red color does not appear in the litmus solution as a result of the base-adsorption of negative electrical soil colloids (which Ramann, in agreement with the opinion expressed by Baumann and Gully insists), but must be considered to express the fact that the soil contains *truly acid reacting substances*. Here, and in what follows, *these substances are to be understood as substances which in solution cause a greater hydrogen-ion concentration than would correspond to the value 10^{-7} [or to a hydrogen-ion exponent (P_H) = 7].*

The author of the present article, collaborating with O. H. Larsen (10), has made investigations of the methods to be used in determining the lime-requirement of the soil. From these it appeared that when a determination

¹¹ In low-bog peat showing a neutral reaction for litmus, the ability to free acids can, as Table IV indicates, be almost as great as in low-bog peat showing acid reaction for litmus. In field-soils many examples (table 6) may be found in which soils with neutral reaction for litmus possess a greater ability to free acids, than soils showing a decided acid reaction for that indicator.

had been made of the reaction of the soil for litmus, and its relation to *Azotobacter*, we possessed a comparatively sure means of determining this requirement. As the results here given, table 6, show there is no definite relation between the reaction of the soil or its power to cause *Azotobacter* development, and its ability to free acid as measured by the acetate method, it is scarcely probable that this method will be of assistance in expressing quantitatively, to a sufficiently correct degree, the lime-requirement of the soil, or as Loew (20, p. 7) in mentioning the acetate method he himself recommends, expresses it—of the need for that amount of lime necessary for neutralizing the soil. Whether the remaining methods which have been suggested (see further, pp. 116, 123) will give better results, must for the present remain doubtful. In any case it is incorrect to assume that the values found by these methods for expressing the ability of the soil to absorb bases also express quantitatively the degree of the lime-requirement of the soil, an assumption, which, as has been mentioned before, is very general.

The variation in the ability of field soils to free acids is found to be very great, just as in the case of humus soils. In fresh moist soils the figures expressing the amount of acid freed per unit of weight vary from 13.18 to 0.18, and when the soil is air-dry from 14.05 to 0.63. As table 6 indicates, the six soils having the greatest power of freeing acid show an acid reaction for litmus, while the nine soils with the smallest power of freeing acid show an alkaline reaction for litmus, but within these broad limits, as I have said before, there is no connection between the reaction of the soil for litmus (or its relation to *Azotobacter*) and its power to free acid from calcium acetate.

The freeing of 1 cc. of 1/10 N acid per gram of dry soil corresponds to the absorption of 2 grams of calcium per kilogram of dry soil. If we estimate that the upper 20 cm. of the field soils (the ordinary ploughed layer) contains 2,400,000 kgm. of air-dry soil per hectare, and assume that bases are absorbed by the soil to the same extent as in the retorts, then this layer has the power to absorb 4800 kgm. of calcium (equal to that amount of calcium found in 12,000 kgm. of carbonate of lime) for every cubic centimeter of 1/10 N acid which 1 gram of soil is able to free from the calcium acetate solution.

The field soil having the greatest power to free acids (no. 761, table 6), will under the given conditions absorb lime in $1.318 \times 12,000 = ca. 16,000$ kgm. of carbonate of lime per hectare (or a corresponding amount of other bases), whereas the soil with the smallest power to free acids (no. 136), will be able to absorb only that amount of lime found in $0.018 \times 12,000 = ca. 200$ kgm. of carbonate of lime per hectare.

If we assume further that raw high-bog peat contains 220,000 kgm. of dry soil, and raw fen peat 400,000 kgm. of dry soil per hectare in a depth of 20 cm. [cf. statements in article (11), table 2, p. 612] the amount of lime (estimated as carbonate) which this layer of peat can absorb per hectare; will in the case of the peat soils mentioned in table 4, series 1, vary from 8820 to 14,960 and 10,400 to 13,600. The ability of the raw peat

TABLE 6
Investigations of the ability of various field soils (mineral soils) to free acid both when fresh and moist, and when air-dry (reaction-time five hours)

MARK	GENERAL CONDITION OF THE SOIL ^a	ACID EFFERVES- CENCE	REACTION	ABSORBACTER VEGETATION	PER CENT OF WATER IN THE SOIL ^b	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID USING						DIFFERENCE BETWEEN MOIST AND AIR-DRY SOIL	
						Fresh, moist soil (10 gm.)			Air-dry soil				
						Per 100 cc. liquid	Per 10 gm. air-dry soil	Per 100 cc. liquid	Per 10 gm. air-dry soil	Per 10 gm. air-dry soil	Per 10 gm. air-dry soil		
761	Loamy, fine-grained dark sandy soil (8)	None	Acid	0	24	3.29 3.37	3.33	13.16	3.54 3.58	3.56	14.05	-0.89	
10	Very light dark sandy soil, poorly humified (2)	-	Acid	0	14	2.51		8.78	2.51		8.78	=0	
682	Light sandy soil, poor in humus (2)	-	Acid	0	13	2.02 2.13	2.08	7.17					
1235	Very light coarse grained sandy soil (1)	-	Slightly acid	0	23	1.70 1.72	1.71	6.66	2.04 2.10	2.07	8.06	-1.40	
901	Clay soil (7)	-	Acid	0	17	1.70 1.74	1.72	6.22					
1259	Sandy loam soil, poor in humus (4)	-	Slightly acid	0	7	1.89 1.82	1.86	6.00	2.01 2.10	2.06	6.65	-0.65	
1260	Loam soil (6)	-	Neutral	4	13	1.77 1.67	1.72	5.93	2.11 2.06	2.09	7.21	-1.28	
775	Light sandy soil, rather rich in humus (4)	-	Slightly acid	0	12	1.63 1.67	1.65	5.63	1.72 1.72	1.72	5.86	-0.23	
321	Good sandy soil (4)	-	Neutral	?	14	1.52 1.50	1.51	5.27	1.80 1.74	1.77	6.18	-0.91	
1240	Loam soil, rather poor in humus (5)	-	Slightly acid	0	10	1.52 1.47	1.50	5.00	1.66 1.66	1.66	5.53	-0.53	
3	Light fine-grained sandy soil (3)	-	Neutral	0-1	12	1.46		4.98	1.78		6.07	-1.09	
13	Good sandy soil, rich in humus (7)	-	Neutral	2	22	1.22		4.69	1.21		4.65	+0.04	

TABLE 6 (Cont.)

MARK	GENERAL CONDITION OF THE SOIL*	ACID EFFERVES- CENCE	REACTION	AUTOMOTIVE VEGETATION	PER CENT OF WATER IN THE SOIL†	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID USING						DIFFERENCE BETWEEN MOIST AND AIR-DRY SOIL	
						Fresh, moist soil (10 gm.)			Air-dry soil†				
						Per 100 cc. liquid	The single retorts	Average	Per 10 gm. air-dry soil	The single retorts	Average	Per 10 gm. air-dry soil	
1242	Light sandy soil, very poor in humus (1)	—	Neutral	1	11	1.29 1.31	1.30		4.38	1.53 1.56	1.55	5.22	-0.84
17	Light sandy soil, poor in humus, marled for 2 years before sample was taken (2)	Rather strong	Neutral	0	7	1.34			4.32	1.46		4.71	-0.39
2662	Clay soil (6)	None	Acid	0	11	1.24			4.18	1.32		4.45	-0.27
745	Light sandy soil (3)	—	Neutral	0	8	1.27	1.27		4.14	1.47	1.46	4.76	-0.62
744	Light sandy soil (3)	—	Neutral—slightly acid	0	9	1.27 1.22	1.25		4.12	1.47 1.42	1.45	4.78	-0.66
727	Clay soil (6)	—	Neutral	3	15	1.16 1.14	1.15		4.06	1.16 1.21	1.19	4.20	-0.14
14	Good sandy soil, rather rich in humus (5)	None	Neutral—slightly alkaline	4	14	0.94			3.28	1.03		3.59	-0.31
786	Sandy loam soil (5)	—	Neutral—slightly alkaline	4	14	0.91 0.94	0.93		3.24	0.99 1.01	1.00	3.49	-0.25
728	Clay soil (7)	—	Neutral	4	13	0.91 0.86	0.89		3.07	0.99 1.01	1.00	3.45	-0.38
18	Light sandy soil, poor in humus (2)	—	Neutral	0	15	0.70			2.47	0.82		2.89	-0.42
7	Clay soil, rich in humus (12)	None—very slight	Alkaline	4	18	0.68			2.46	1.11		4.01	-1.55
16	Very light sandy soil, poor in humus (1)	None	Neutral	0	6	0.76			2.43	0.83		2.65	-0.22
9	Loam soil (6)	Very slight	Slightly alkaline	4	15	0.62			2.19	0.68		2.40	-0.21

TABLE 6 (Cont.)

MARK	GENERAL CONDITION OF THE SOIL*	ACID EFFERVESCENCE	REACTION	AEROBACTER VEGETATION	PER CENT OF WATER IN THE SOIL†	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID USING				DIFFERENCE BETWEEN MOIST AND AIR-DRY SOIL	
						Fresh, moist soil (10 gm.)		Air-dry soil			
						Per 100 cc. liquid	Per 10 gm. air-dry soil	Per 100 cc. liquid	Per 10 gm. air-dry soil		
11	Light sandy soil (3)	None—very slight	Slightly alkaline	4	90.64	2.11	0.77	2.54	-0.43		
773	Dark sandy soil, rather rich in humus (5)	None	Slightly alkaline	4	130.56 0.56	0.56	1.93 0.81	0.76	2.62	-0.69	
8	Clay soil (7)	Very slight	Alkaline	4	150.52	1.84	0.52	1.84	± 0		
790	Clay soil, rich in humus (10)	Very slight	Slightly alkaline	4	240.46 0.41	0.44	1.74 0.76	0.76	3.00	-1.26	
800	Dark sandy soil, rich in humus (7)	Slight	Alkaline	4	310.42 0.38	0.40	1.74 0.56	0.56	2.43	-0.69	
a	Clay soil, rich in humus (10)	Strong	Alkaline	4	60.44	1.40					
1262	Sandy loam soil, poor in humus (4)	Slight	Alkaline	4	70.28 0.28	0.28	0.90 0.60	0.62 0.61	1.97	-1.07	
736	Clay soil, poor in humus (6)	None	Alkaline	4	150.07 0.02	0.05	0.18 0.23	0.23	0.81	-0.63	

* The figures in parentheses indicate the degree of heaviness of the soil.

† In the case of field soils the percentage of water is determined by air-drying.

soils to absorb bases (estimated per cubic measure) is not greater than that of several field soils.

The absorption of basic substances depends naturally on their being rendered soluble. Carbonate of lime is, for the most part, held in solution by the soil water, as bicarbonate of lime. The lime in this compound is doubtless absorbed very quickly by the soil—presumably more quickly and completely than when it appears as an acetate. Therefore the rapidity with which the lime is absorbed by "non-base saturate" soil is more than probably dependent on how soon it is rendered soluble. This, in turn, depends on the amount of moisture in the soil which in a given time has an opportunity of affecting the lime.

The author has previously had an opportunity of making investigations in connection with field experiments under the leadership of F. Kölpin Ravn,

(22) at the Askov Experiment Station, as to the use of lime as a preventive for club-root (*Plasmodephora brassicae*). In the loam soils used it was found, that lime may be absorbed very quickly from carbonate of lime, even in soils showing a neutral reaction for litmus. In the soils which in 1906 were treated with *ca.* 4000 kgm. of CaCO₃ per hectare no carbonates could be found 4½ years later under an examination, in which the acid test was used (sprinkling the soil with diluted HCl). Even when 12,000 kgm. of CaCO₃ were used per hectare, after the same length of time had elapsed, only a very slight effervescence was apparent in three of the four replicate plots. By the acid test made in 1911, two of the four replicate plots, which for four years (1902-1906) received an annual dose of *ca.* 3000 kgm. of CaCO₃ per hectare—12,000 kgm. in all—showed only a slight effervescence when acid was added, while specimens from the two remaining plots showed no effervescence whatsoever. As this acid test indicates the presence of very small amounts of carbonates in the soil, and the loss of carbonate of lime by washing-out during these few years cannot have been very great, these results may be considered an indication that the amount of calcium absorbed by the soil from the calcium carbonate, may attain quite a considerable dimension within a comparatively short period of time. In regard to the influence of this condition upon the power of carbonate of lime to change the reaction of the soil, see the paper (22) mentioned above.

Based on the results mentioned before, the following suggestion for a method for determining the power of the soil to free acids from calcium acetate is made.

Into a retort with a capacity of at least 400 cc., measure off 300 cc. of calcium acetate solution (10 grams of calcium acetate to 100 cc. of distilled water). Place in the retort fresh moist soil specimens, taking of pure humus soils (i.e., unmixed with mineral matter) as much of the finely divided mass as would correspond to about 0.5 gram of oven-dry soil, and of mineral soils (field soils) as much as would correspond to about 10 grams of air-dry soil. The reaction-time must be at least ten hours.¹² During the first and last part of the reaction-time the retorts are to be shaken repeatedly. As to methods of procedure for filtering and titrating, see the report, page 123.

When especially complete and accurate information as to the ability of the soil to free acid is wanted, it will be necessary, as we have mentioned before, to vary the relation between the amount of soil and the amount of solution. Of peat soils two different quantities might be used, corresponding to 0.5 gram and 2 grams of oven-dry peat, and of mineral soils two quantities corresponding to 10 grams and 20 grams of air-dry soil (to 300 cc. of acetate solution). As the results of the duplicate determinations always agree, always

¹² Even though a reaction-time of ten hours based on the investigations already made, might seem sufficient to attain the maximum amount of acid freed, yet in order to arrange laboratory work in a practical way, it is advisable to use a longer reaction-time. The soil might be placed in the retorts about 2 hours before the laboratory is closed. The retorts stand over night, and a filtration is made the next morning.

supposing that no serious errors have been made, a repetition of the investigation may be omitted, especially when two different quantities of soil have been used, for the results here obtained control each other to a certain extent.

The results of the investigations are given, for humus soils in cubic centimeters of 1/10 N acid per gram of oven-dry soil, and for mineral soils in cubic centimeters of 1/10 N acid per gram (or 10 grams) of air-dry soil, for in the latter case the amount of water hygroscopically bound is so small that it may be omitted in calculating the results.

3. Determination of the ability of the soil to free acid from calcium-carbonate by means of the Tacke-Süchting method

The principle underlying this method is mentioned on page 116. The apparatus used, shown in figure 3, page 139, corresponds very closely to that used by Tacke and Süchting [and described by Tacke in paper (30)]. The method used in making the determinations, which also for the most part corresponds to that used by the two scientists, was as follows.

Moist soil, finely minced in a meat-chopper, is placed in a reaction-retort (*e*) and from the hydrogen-generating apparatus (*b*), which in order to increase the hydrogen pressure is equipped with a manometer-tube (*a*) with mercury, hydrogen is led through a side tube into the reaction retort. Before the hydrogen reaches this, it has passed through two washing bottles (*c* and *d*) containing strong sulfuric acid and strong sodium hydroxide, respectively. The combined hydrogen and carbonic acid current now passes from the reaction-retort through a Pettenkofer absorption tube (*f*), containing 100 cc. of diluted sodium hydroxide (*ca.* 1/10 N), and is finally led off into a beaker (*g*) half-full of water. The speed of the hydrogen current is regulated by means of a stop-cock (*k*) on the side-tube of the hydrogen-generating apparatus. The agitator, with a mercury stopper, is driven by an electro-motor.

The peat soil, weighed off, is placed in the reaction-retort. This is supplied with water [Tacke (30, p. 121), mentions that the retorts are half filled with water], and a not too large surplus of carbonate of lime added. After an elongation of the tube projecting from the reaction-retort has been placed under water in a beaker, a hydrogen current (*ca.* two bubbles per second) is passed through the apparatus in order to drive off the carbonic acid generated by the action of the carbonate of lime on the peat soils. (This according to Tacke and Süchting will be accomplished after two hours.) The absorption tube is now nearly filled with *ca* 1/10 N sodium hydroxide, and connected with the apparatus. Now about 50 cc. of 20 per cent HCl is poured through the tube (*h*), which has been connected with a separatory, and the carbonic acid freed from the remaining carbonate of lime is conducted by help of the hydrogen current, and under continual agitating through the absorption-tube. (According to Tacke and Süchting a hydrogen current for an hour is sufficient to drive off the entire amount of CO₂).

While Tacke and Süchting have determined the content of CO₂ in the receiver by titrating according to Cl. Winkler's method (adding pure barium chloride, and titrating with 1/5 or 1/10 N HCl, using phenolphthalein as an indicator), here the following method, which is very easy to work with, has been used.

From the 100 cc. of sodium hydroxide in the Pettenkofer tube, take 40 cc. and to it add 25 cc. of a 5 per cent barium chloride solution. After the pre-

cipitated barium carbonate has been removed by filtering, 50 cc. of the filtrate should be taken and this portion titrated with HCl, using methyl-orange as an indicator.

By multiplying the value found with $\frac{65 \times 100}{50 \times 40} = 3.25$ the total amount of NaOH in the Pettenkofer tube may be estimated, and by subtracting this amount from the amount of NaOH originally present, we have an expression for the amount of CO₂ freed. The amount of NaOH in the sodium hydroxide solution in its original form is determined in a similar fashion. One cubic centimeter 1/10 N NaOH corresponds to 0.0022 gram of CO₂.

TABLE 7
Influence of the amount of peat used upon the ability to free CO₂

AMOUNT OF PEAT	AMOUNT OF WATER: IN CUBIC CENTIMETERS	AMOUNT OF CaCO ₃ USED: IN GRAMS	GRAMS OF CO ₂					GRAMS OF CO ₂ PER 10 GRAMS PEAT	
			Single determinations						
			a	b	c	d	e		
<i>High-bog peat A (stirred for 3 hours)</i>									
10	1.51	415	0.1747	0.0151	0.0138	0.0162	0.0160	0.0162	0.0155
15	2.27	415	0.2005	0.0243	0.0305				0.0274
20	3.02	415	0.1959	0.0384	0.0385				0.0385
30	4.53	415	0.2199	0.0589	0.0539				0.0564
<i>High-bog peat B (stirred for 3 hours)</i>									
10	1.47	200	0.1766	0.0241	0.0225				0.0233
20	2.94	200	0.2115	0.0523	0.0459				0.0491

The investigations of the Tacke-Süchting method aim to throw light on the following questions:

1. What influence may varying the amount of peat used have on freeing CO₂, the amount of water being kept constant?
2. What influence may varying the amount of water used have on freeing CO₂, the amount of peat being kept constant?
3. The influence of the reaction-time on the freeing of CO₂.

The results of the investigations may be seen in tables 7 and 8.

If we first turn to table 7, we see that the results give no indication that an increase of the amount of peat has relatively reduced the CO₂ freed. Quite the contrary is true, for in high-bog peat specimen A this is smallest, when the smallest amount of peat is used. In high-bog peat specimen B approximately as large an amount of CO₂ is freed per weight unit of peat when a small amount of peat is used, as when a large amount is used.

Table 8 gives the results of investigations as to the influence which varying the amount of water and the reaction-time may exert upon freeing CO₂,

and here there is a much greater result from the factors tested than in the investigations mentioned before.

When the difference between the amounts of peat and of water is relatively small, the amount of CO_2 freed in the same period is greater than when the difference is large. Moreover, we find that the reaction-time exerts a very great influence on the amount of CO_2 freed, and that this influence is especially apparent when the largest amount of water is used. In high-bog peat specimen A, for example, more CO_2 is freed when the smallest amount of water is used with a reaction-time of three hours, than when the largest

TABLE 8
Influence of the reaction-time and the amount of water used upon the ability to free CO_2

REACTION TIME IN HOURS	RELATION BETWEEN AMOUNTS OF SOIL AND WATER	OVEN-DRY PEAT USED: IN GRAMS	AMOUNT OF CaCO_3 USED: IN GRAMS	GRAMS OF CO_2					Average	
				Single determinations						
				a	b	c	d	e		
<i>High-bog peat A</i>										
3	10: 200	1.51	0.179	0.0255	0.0225	0.0252			0.0244	
4	10: 200	1.51	0.191	0.0263	0.0265				0.0264	
1	10: 415	1.51	0.164	0.0067	0.0056				0.0062	
2	10: 415	1.51	0.166	0.0124	0.0128				0.0126	
3	10: 415	1.51	0.175	0.0162	0.0151	0.0138	0.0162	0.0160	0.0155	
4	10: 415	1.51	0.165	0.0198	0.0192				0.0195	
6	10: 415	1.51	0.200	0.0236	0.0223				0.0230	
8	10: 415	1.51	0.211	0.0252	0.0211				(?)	
<i>High-bog peat B</i>										
2	10.3: 100	1.51	0.202	0.0271	0.0226				0.0249	
3	10.3: 100	1.51	0.201	0.0255	0.0248				0.0253	
2	10.3: 200	1.51	0.169	0.0225	0.0188				0.0207	
3	10.3: 200	1.51	0.178	0.0270	0.0263				0.0267	
4	10.3: 200	1.51	0.185	0.0273	0.0283				0.0278	
6	10.3: 200	1.51	0.194	0.0314	0.0318				0.0316	

amount of water is used with a six-hour reaction-time. In the latter case CO_2 has not totally ceased to generate even when six hours have elapsed.

The investigations made with high-bog peat specimen B, in which the amount of water used was much smaller, seem also to indicate that CO_2 is freed most quickly when the smallest amount of water is used, even though this result does not appear very plainly because of poor agreement in the duplicate determinations. In a reaction-time of three hours, and when the largest amount of water is used (200 cc.), we have in this experiment just as great an amount of CO_2 as by using the smallest amount of water (100 cc.). But also, when so moderate an amount of water as 200 cc. is used, a long time

must elapse before the reaction between peat and lime ceases, for even after four hours CO_2 is still being generated.

In the investigations made by Sückting (27), using two different specimens of high-bog peat, a two-hour reaction-time has proved quite sufficient to bring the reaction to an end (27, p. 42). This result is opposed to the results of the investigations which we have here. For instance, from table 8 it appears that in high-bog peat nearly twice as much CO_2 is freed after six hours have passed, as after two hours, and that the amount of CO_2 freed increases gradually with the length of the reaction-time. As the amount of water used, as well as the reaction-time, determines the amount of CO_2 freed, it must be admitted that in this investigation, using the Tacke-Sückting method, it has been impossible to obtain an exact expression of the ability of the soil to free CO_2 from CaCO_3 . The author is unable to explain the great difference between Sückting's results and those given here. However, for the

TABLE 9
Investigations of the time required for airing with hydrogen

PEAT USED	RELATION BETWEEN AMOUNTS OF PEAT AND WATER USED	AMOUNT OF CaCO_3 USED: IN GRAMS	GRAMS OF CO_2 AFTER AIRING WITH HYDROGEN FOR			DIFFERENCE BETWEEN AIRING FOR 1 HOUR AND 3 HOURS		DIFFERENCE BETWEEN AIRING FOR 3 HOURS AND 5 HOURS	
			1 Hour	3 Hours	5 Hours	Grams	Percentage of CO_2 found after 1 hour	Grams	Percentage of CO_2 found after 3 hours
High-bog peat A....	10: 415	0.1741	0.0435	0.0604		0.0169	38.9		
High-bog peat A....	10: 415	0.1778		0.0618	0.0673			0.0055	8.9
High-bog peat B....	10.3: 200	0.1780		0.0520	0.0541			0.0021	4.0

present, he will limit himself to stating, that the two-hour reaction-time suggested by Tacke and Sückting is in any case far too short to bring the reaction between high-bog peat and carbonate of lime to a complete close, and that in the investigations made here, as well as in those applied to the calcium acetate method, before mentioned, the length of the reaction-time to a far greater extent than the amount of peat used appears to determine the amount of acid freed per weight unit of peat.

Nor has it been possible to confirm Sückting's results in regard to the time necessary for bringing into the receiver the entire amount of CO_2 freed by the addition of HCl. According to Sückting's investigations, after hydrogen has been conducted through the apparatus for an hour no more CO_2 passes into the receiver, while according to our investigations no such constant results have been attained. By using a comparatively large amount of water, the difference between the amount of CO_2 collected after hydrogen has been conducted for one and three hours is so great, that it must to a large extent be due to the fact that the first-named period was insufficient for driving the

amount of CO_2 freed from calcium carbonate into the receiver, whereas the comparatively small differences in the amount of CO_2 after hydrogen has been conducted through the apparatus for three and five hours, may possibly be explained by internal reactions in the humus materials themselves.

In all the investigations referred to in tables 7 and 8, hydrogen has been conducted through the apparatus not only one, but three hours after hydrochloric acid has been added.

As an objection to the principle underlying the acetate method, Sückting (27, p. 23), criticizing an experiment made by Berthelot to obtain an expression for the acidity of the soil by determining the amount of acetic acid which a certain quantity of soil when heated with a solution of potassium acetate is able to free from that salt, emphasizes that aside from other weaknesses, by this method one is able only to determine the amount of soil acids

TABLE 10
Comparison between the amounts of acid freed, using the Tacke-Sückting method and the calcium acetate method

PEAT SOIL	AMOUNT OF ACID FREED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID PER 1 GRAM OF OVEN-DRY PEAT					
	Tacke-Sückting method (Reaction-time 3 hours)	Calcium acetate method (6 grams moist peat to 300 cubic centimeters liquid)	Relation between amount of moist peat* and water			
			10:200	10:415	3 hours	5 hours
High-bog peat A.....	7.3	4.7	14.0	14.1		
High-bog peat B.....	8.0		10.0		10.6	

* Each peat sample contained 1.51 gram of oven-dry substance in 10 grams of moist peat.

which are stronger than acetic acid, and therefore the method gives us no accurate expression for the acidity of the soil.

In table 10 a comparison has been made between the calcium acetate method and the Tacke-Sückting method applied to investigations in freeing acid in high-bog peat specimens A and B (tables 7 and 8).

Although in the Tacke-Sückting method a three-hour instead of a two-hour reaction-time has been used, which must have increased the amount of CO_2 freed considerably (table 8), yet the method in every instance gives distinctly lower results than the calcium acetate method. The difference in results from the two methods is especially apparent in high-bog peat specimen A. Here the Tacke-Sückting method gives only about one-half as large results as the calcium acetate method when the smallest amount of water is used, and about one-third as large when the largest amount of water is used.

If, as does Sückting, we would take for granted, that the amount of calcium absorbed expresses the acidity of the soil, the figures appearing when this last-named method is used must more nearly approach the absolute value for this acidity than the figures appearing from the first method. Moreover, as the results from the acetate method show that there is far better concordance in the results from the duplicate determinations, it seems as if this method should be preferred to the Tacke-Sückting method, over which it has the additional advantage that it is quicker and more easy to work with.

II. QUALITATIVE DETERMINATION OF THE REACTION OF THE SOIL

1. Methods for making a qualitative determination of the reaction of the soil

In making a qualitative determination of the reaction of the soil, litmus paper is most frequently used, and the method is generally as follows [see, for instance, M. Weibull (33)]. A small portion of the soil sample is placed in a dish and moistened with enough water to give it, when stirred, a porridge-like consistency. A strip of red and a strip of blue litmus paper are now placed over the soil in such a way, that the paper at every point comes in contact with the soil particles. A few hours later the color changes in the litmus paper are observed and noted.

In making a qualitative determination of the soil reaction the author of this paper has used a neutral litmus solution. His method is described in detail in a previous paper (10, p. 431).

In 1908 Baumann and Gully (3) published a method for making a qualitative acidity determination based on the ability of free acids to free iodine from a solution of potassium iodide and potassium iodate.

The method is as follows.

Two grams of potassium iodide and 0.1 gram of potassium iodate are dissolved in 100 cc. of distilled water. One gram of peat soil (if mineral soil, a slightly larger portion) is placed in the retort, which is shaken frequently, and fifteen minutes later filtered. A few drops of the filtrate are placed in a thin starch water and the intensity of the blue color noted.

Recently Daikuahara (12) proposed the following method for making a qualitative determination of the acidity of the soil.¹²

Place 5 grams of soil in a test tube and add drop by drop a 10 per cent potassium nitrite solution until the soil is well moistened. The tube is corked with a wad of cotton, from which a strip of potassium iodide starch paper reaches down into the tube. After a short time the intensity of the blue color of the paper gives an indication as to the degree of the acidity of the soil. The potassium nitrite used must be chemically pure, and above all must not contain any potassium carbonate. Potassium nitrite is considered better in such investigations than sodium nitrite.

The results obtained from this method, as well as those from the Baumann-Gully method, can give only indications as to the intensity of the acidity of the soil, but no indication, as the litmus method to some extent does, as to the alkalinity of the soil.

¹² This method is a modification of a method previously proposed by Oscar Loew (20).

2. Comparison of various methods for making a qualitative determination of the reaction of the soil

Considering the important part which a qualitative determination of the reaction and basicity of the soil has played in later years in soil investigations, a comparison of the most important methods suggested would seem to be of interest in this connection, and therefore the investigations referred to in table 11 have been made.

In these investigations the following methods have been used.

Litmus method (litmus test), with the use of both litmus solution and litmus paper. In investigating the question of the extent to which air-drying the soil affects its reaction, litmus solution being used, a determination was made of soil samples both in that condition of moisture which was present at the beginning of the experiment, and air-dry. Two equally large soil portions were simultaneously weighed off. One of these was placed in a test tube which was at once corked, and the other spread out on a piece of paper to become air-dry. After drying, this portion was likewise placed in a test tube and to each tube 20 cc. of neutral litmus solution added. Of mineral soil a portion was used corresponding to 5 grams of air-dry soil, and of fresh moist peat as much as would correspond to 1 gram of oven-dry peat.

Nitrite method. The one I have used is closely related to the one suggested by Daikuahara. However, in place of potassium nitrite, sodium nitrite is used, because neutral potassium nitrite could not be procured. A portion of the fresh moist soil corresponding to about 5 grams of air-dry soil was used. The reagent paper was moistened with distilled water before being submerged in the glass. It has been found important not to use more nitrite solution than is just sufficient to moisten the soil sample. In the presence of a greater amount of moisture the reaction is often irregular.

Iodine method (according to Baumann and Gully). For this method, see above. As the prepared solution of potassium iodide and potassium iodate showed a slightly acid reaction for litmus paper, and was distinctly yellow in color, it was neutralized before use with a very thin sodium hydroxide solution, and the color finally entirely removed by adding, drop by drop, a very thin sodium hyposulfite solution. With 100 cc. of the named solution of potassium iodide, and potassium iodate, a portion of mineral soil corresponding to 3 grams of air-dry soil was used. Besides noting the degree of the blue color in the starch water, the degree of the yellow color in the filtrate itself was noted, for this reaction seemed to give better and more complete information as to the acidity of the soil, than the starch water reaction. The degree of this last depends to a certain extent on the concentration of the starch water used, and it was found necessary to use a rather strongly diluted starch water. Observation of the blue color must be made immediately after the filtrate has been dropped, as the starch solution will, in any case after a short time, assume a blue color (due to the sensitiveness of the reagent to CO_2). Only

two shades of blue can be distinguished with positive certainty—light blue and blue.

In addition to these indicators, experiments have been made with *p-nitrophenol*. This substance, tested by S. P. L. Sørensen (23) in measuring hydrogen-ion concentration in approximately neutral stuffs, has proved to be a splendid indicator.¹⁴ Of this indicator, which, as far as is known, has not previously been used in making soil-reaction determinations, a solution of the following composition was prepared: 0.4 gram *p*-nitrophenol, 60 cc. of alcohol, and 940 cc. of distilled water. Twenty drops of the solution were used to 20 cc. of distilled water, which at once assumed a pale yellowish color. In this test the mineral soils used were always in an air-dry condition, and 5 grams taken for each determination. Peat soils were used in their original moist condition, and in the amount mentioned above. The soil and the solution were well shaken together and the observations of the color of the solution made the following day. Decidedly acid soils render the solution colorless, while slightly acid, neutral and alkaline soils color it yellow (varying from a very pale to a greenish yellow).

In connection with these qualitative investigations, quantitative determinations have been made, based on the calcium acetate method, and the Daikuvara potassium chloride method. In the former a portion of soil corresponding to 9 grams of air-dry field soil, or 0.5 gram of oven-dry peat soil, to 300 cc. of calcium acetate solution is used. The mixture stood for *ca.* eighteen hours, was shaken from time to time and titrated according to the method mentioned before. In the potassium chloride method of field soils a 100 gram portion of the fresh soil specimen, and of peat soils a portion corresponding to 10 grams of oven-dry peat, was weighed off to 250 cc. 1/1 N potassium chloride solution. The mixture stood for five days, being shaken from time to time. Half of the amount of fluid present (= 125 cc. of potassium chloride solution plus half of the moisture found in the soil added), corresponding to half of the portion of soil used, is titrated (after having been filtered, and the free CO₂ removed by hasty boiling), with 1/10 N NaOH, phenolphthalein being used as the indicator. According to Daikuvara the content of acid found is multiplied by the factor 3, and thereby, as has been mentioned before, an approximately correct expression for the absolute acidity of the soil should be obtained (see further, page 117).

As is mentioned on page 117, Daikuvara states that the acidity of mineral soils is for the most part determined by the presence of acid aluminum—or (in certain instances), iron-compounds, and that the amount of these found in the potassium chloride solution corresponds very closely to the amount of NaOH used in titration.

¹⁴ Both *p*-nitrophenol and litmus belong to that group of indicators which S. P. L. Sørensen calls "indicators of the phosphate mixtures," and which are characterized by the fact that their turning-point is very close to the true neutral point ($P_H = 7$). Litmus spans the hydrogen-ion domain: $P_H = ca. 4.5$ to 8.3 and *p*-nitrophenol the hydrogen-ion domain $P_H = 5$ to 7.

TABLE 11
Comparison of various methods for determining the reaction of the soil

718	Gray sandy soil, slightly mixed with peat (2)	85.2	None	0	Strong acid	Acid	Acid	Very strong	Strong yellow	Blue	Colorless	3.14	104.7
1	Clay soil; poor in humus (5)	86.0	Very slight	0	Strong acid	Acid	Acid	Very strong	Yellow	Blue	Colorless	1.72	57.3
2	Clay soil; poor in humus (6)	86	Very slight	0	Strong acid	Strong acid	Acid	Very strong	Yellow	Very pale yellow	Colorless	1.44	48.0
0997	Good sandy soil (4)	90.0	None	0	Acid	Slightly acid	Acid	Very strong	Yellow	Light blue	Colorless	2.16	72.0
0105	Light sandy soil (3)	89.6	None	0	Acid	Strong acid	Acid	Very strong	Strong yellow	Blue	Colorless	3.02	100.7
977	Loam soil, rather poor in humus (5)	94.8	None	0	Acid	Slightly acid	Acid	Very strong	Strong yellow	Blue	Colorless	2.84	94.7
0557	Good sandy soil (4)	90.2	None	0	Acid	Slightly acid	Acid	Very strong	Strong yellow	Blue	Colorless	1.88	62.7
0228	Fine sandy soil, poor in humus (4)	91.0	None	0	Acid	Acid	Acid	Very strong	Strong yellow	Blue	Colorless	1.64	54.7
0667	Good sandy soil, rather rich in humus (5)	84.0	None	0	Acid	Acid	Acid	Very strong	Pale yellow	Light blue	Colorless	2.70	90.0
062	Light fine sandy soil, rather poor in humus (3)	92.0	None	0	Acid	Strongly acid	Slightly acid	Very strong	Pale yellow	Blue	Colorless	1.40	46.7
0983	Clay soil (8)	94.6	None	0	Acid	Acid	Acid	Very strong	Pale yellow	Light blue	Colorless	1.52	50.7
332	Light sandy soil, poor in humus(2)	91.8	None	0	Acid	Acid	Acid	Very strong	Pale yellow	Blue	Colorless	1.58	52.7
284	Sandy loam soil (5)	93.8	None	0	Acid	Slightly acid	Acid	Very strong	Very pale yellow	Light blue	Colorless	1.30	43.3
946	Light, rather coarse sandy soil (2)	93.8	None	0	Slightly acid	Slightly acid	Acid	Very strong	Yellow	Blue	Colorless	1.38	46.0
1178	Loam soil (6)	92.0	None	0	acid	Slightly acid	Acid	Very strong	Strong yellow	Blue	Colorless	1.52	50.7
127	Loam soil (6)	94.8	None	0	Slightly acid	Slightly acid	Acid	Very strong	Rather strong-slight	Yellow	Colorless	1.59	53.0
1158	Loam soil, poor in humus (5)	93.8	None	0	Slightly acid	Acid	Acid	Very strong	—very pale yellow	Blue	Colorless	1.60	53.3

TABLE 11 (Cont.)

SOIL SAMPLE NUMBER	ORDINARY CONDITION OF THE SOIL	REACTION FOR LITHIUM SOLUTION		REACTION FOR LITHIUM IN Mixture PAPER (EOL FOR-NITRO-POTASSIUM CHLORIDE)		REACTION FOR LITHIUM IN Mixture PAPER (EOL FOR-NITRO-PHENOL)		ACID TESTED, EXPRESSED IN CUDIC CHARTERS 1/10 N ACID	
		ACID DIFFERENCE	PER CENT DRY MATTER IN THE SOIL	0	0	0	0	0	0
3114	Clay soil, poor in humus (6)	None	96.6	Slightly acid	Slightly acid	Slightly acid	Acid	Very strong	Very pale Yellow
4042	Good sandy soil (4)	None	90.8	Slightly acid	Slightly acid	Slightly acid	Slightly acid	Strong	Yellow
4079	Sandy loam soil, rather poor in humus (4)	None	85.0	Slightly acid	Slightly acid	Slightly acid	Acid	Strong	Blue
4041	Clay soil (7)	None	85.6	Slightly acid	Slightly acid	Slightly acid	Acid	Strong	Blue
4350	Light sandy soil (3)	None	89.0	Slightly acid	Slightly acid	Slightly acid	Acid	Very strong	Blue
4323	Light sandy soil (3)	None	88.4	Slightly acid	Slightly acid	Slightly acid	Neutral—slightly acid	Very strong	Blue
1944	Light rather coarse sandy soil (2)	None	97.6	Neutral—slightly acid	Neutral—slightly acid	Neutral—slightly acid	Neutral—slightly acid	Strong	Blue
1919	Coarse sandy soil, very poor in humus (2)	None	96.0	Neutral—slightly acid	Neutral—slightly acid	Neutral—slightly acid	Neutral—slightly acid	Strong	Blue

1. Field Soils

3114	Clay soil, poor in humus (6)	None	96.6	0	0	0	0	0.92	30.7	0.30	1.9	None	1: 16.2
4042	Good sandy soil (4)	None	90.8	0	0	0	0	1.52	30.7	0.25	1.7	None	1: 29.8
4079	Sandy loam soil, rather poor in humus (4)	None	85.0	0	0	0	0	1.26	42.0	0.29	2.0	None	1: 21
4041	Clay soil (7)	None	85.6	0	0	0	0	1.44	48.0	0.15	1.1	None	1: 43.6
4350	Light sandy soil (3)	None	89.0	0	0	0	0	1.70	36.7	0.53	3.6	None	1: 15.8
4323	Light sandy soil (3)	None	88.4	0	0	0	0	1.68	36.0	0.24	1.6	None	1: 35.0
1944	Light rather coarse sandy soil (2)	None	97.6	0	0	0	0	1.28	42.7	0.86	5.3	Small	1: 8.1
1919	Coarse sandy soil, very poor in humus (2)	None	96.0	0	0	0	0	0.67	22.3	0.48	2.9	Small	1: 7.7

2982	Good sandy soil (4)	96.6	None	0	Neutral—slightly acid	Slightly acid	Slight	Yellow	Light blue	Very pale yellow	1.90	63.3	0.75	1.6	None	1: 39.6	
3073	Clay soil, poor in humus (6)	95.4	None	0	Neutral—slightly acid	Neutral—slightly acid	Slightly acid	Yellow	Light blue	Very pale yellow	0.98	32.5	0.29	1.8	None	1: 18.1	
2937	Good sandy soil (4)	93.0	None	0	Neutral—slightly acid	Neutral	Neutral	Pale yellow	Colorless	Very pale yellow	1.09	36.3	0.14	0.9	None	1: 40	
4027	Clay soil, poor in humus (6)	90.0	None	0	Neutral—slightly acid	Neutral—slightly acid	Neutral—slightly acid	Very strong—slight	Blue	Very pale yellow	1.48	49.3	0.15	1.0	None	1: 49.3	
4044	Good sandy soil, rather rich in humus (5)	88.3	None	0	Neutral—slightly acid	Neutral	Neutral	Very strong	Colorless	Colorless	Very pale yellow	1.50	50.0	0.09	0.6	None	1: 83.3
4333	Light sandy soil (3)	88.2	None	0	Neutral—slightly acid	Neutral	Neutral	Very strong	Colorless	Colorless	Very pale yellow	1.56	52.0	0.25	1.7	None	1: 30.6
4376	Clay soil, rather rich in humus (9)	86.4	None	0	Neutral—slightly acid	Neutral—slightly acid	Slightly acid	Very pale yellow	Colorless	Very pale yellow	1.28	42.7	0.22	1.5	None	1: 28.5	
2951	Good sandy soil, rich in humus (6)	91.3	None	0	Neutral—slightly acid	Neutral	Neutral	Strong—slightly alkaline	Colorless	Colorless	Very pale yellow	2.11	70.3	0.12	0.8	None	1: 87.9
3074	Loam soil (6)	94.5	None	0	Neutral	Neutral	Neutral	Strong—slightly alkaline	Colorless	Colorless	Very pale yellow	0.84	28.0	0.10	0.6	None	1: 46.7
27	Light sandy soil (3)	93.4	None	0	Neutral	Neutral	Neutral	Very pale yellow	Colorless	Colorless	Very pale yellow	0.80	26.7	0.10	0.6	None	1: 44.5
4088	Sandy loam soil, poor in humus (4)	84.4	None	0	Neutral	Neutral	Neutral	Very pale yellow	Colorless	Colorless	Very pale yellow	1.08	36.0	0.13	0.6	None	1: 60.0
4429	Sandy loam soil (5)	87.2	None	0	Neutral	Neutral	Neutral	Rather strong	Colorless	Colorless	Very pale yellow	0.92	30.7	0.13	1.0	None	1: 30.7
4362	Good sandy soil, rich in humus (6)	86.2	None	0	Neutral	Neutral	Neutral	Rather strong	Colorless	Colorless	Pale yellow	1.20	40.0	0.16	1.0	None	1: 40.0
4011	Sandy loam soil (5)	85.0	None	4	Neutral	Neutral	Neutral	Slight	Colorless	Colorless	Pale yellow	0.96	32.0	0.15	1.1	None	1: 29.1
4045	Sandy loam soil, poor in humus (4)	88.0	None	4	Neutral	Neutral	Neutral	Rather strong, slight	Colorless	Colorless	Pale yellow	0.68	22.7	0.08	0.5	None	1: 45.4

TABLE II (Cont.)

SOIL SAMPLE NUMBER	ORDINARY CONDITION OF THE SOIL	ACID EXTRACT VESICULARITY		REACTION FOR LITMUS SOLUTION		REACTION FOR LITMUS IN PAPER (SOIL PONDAGE)		REACTION FOR LITMUS IN MIXTURE OF SOIL AND POTASSIUM CHLORIDE SOLUTION		REACTION FOR POTASSIUM IODIDE AND POTASSIUM IODATE (BALTANN® AND GULLY'S METHOD)		REACTION FOR p-NITRO-PHENOL (BALTANN® AND GULLY'S METHOD)		ACID FREED, EXPRESSED IN CUBIC CENTIMETERS /1/10 N ACID		RELATION BETWEEN THE AMOUNT OF ACID USED AND THE POTASSIUM HYDRATE PREPARED WHEN KCl IS ADDED TO THE SOIL	
		PER CENT DRY MATTER IN THE SOIL	PER CENT DRY MATTER IN THE SOIL	NEUTRAL	ACID	NEUTRAL	ACID	NEUTRAL	ACID	NEUTRAL	ACID	NEUTRAL	NEUTRAL	ACID	NEUTRAL	ACID	NEUTRAL
2932	Good dark sandy soil, very rich in humus (8)	75.0	None	4	Neutral	Slightly alkaline	Neutral	Colorless	Pale yellow	1.38	46.0	0.14	1.1	None	1: 41.8		
2969	Good sandy soil (4)	91.2	None	4	Neutral	Neutral—slightly acid	Neutral	Colorless	Very pale yellow	0.80	26.7	0.05	0.3	None	1: 89.0		
2923	Sandy loam soil (5)	91.8	None	4	Neutral	Neutral	Neutral	Colorless	Very pale yellow	1.05	35.0	0.14	0.9	None	1: 38.9		
3064	Loam soil, poor in humus (5)	92.6	Very slight	4	Neutral	Neutral—slightly alkaline	Slightly alkaline	Colorless	Pale yellow	0.61	20.3	0.10	0.6	None	1: 33.8		
3081	Sandy loam soil (5)	93.2	None	4	Neutral	Neutral—slightly alkaline	Slightly alkaline	Colorless	Pale yellow	0.52	17.3	0.10	0.6	None	1: 28.8		
2922	Sandy loam soil (5)	90.0	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.01	33.7	0.14	0.9	None	1: 37.4		
3058	Clay soil, poor in humus (6)	97.2	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.24	41.3	0.10	0.6	None	1: 68.8		
3141	Clay soil (9)	83.0	None	4	Neutral	Neutral—slightly alkaline	Neutral	Colorless	Pale yellow	1.18	39.3	0.10	0.7	None	1: 56.1		

1. Field Soils											
2932	Good dark sandy soil, very rich in humus (8)	75.0	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.38	46.0
2969	Good sandy soil (4)	91.2	None	4	Neutral	Neutral	Neutral	Colorless	Very pale yellow	0.80	26.7
2923	Sandy loam soil (5)	91.8	None	4	Neutral	Neutral	Neutral	Colorless	Very pale yellow	1.05	35.0
3064	Loam soil, poor in humus (5)	92.6	Very slight	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	0.61	20.3
3081	Sandy loam soil (5)	93.2	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	0.52	17.3
2922	Sandy loam soil (5)	90.0	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.01	33.7
3058	Clay soil, poor in humus (6)	97.2	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.24	41.3
3141	Clay soil (9)	83.0	None	4	Neutral	Neutral	Neutral	Colorless	Pale yellow	1.18	39.3

160	Good sandy soil (4)	92.0	None		4	Neutral	Neutral	Neutral	Very slight, slight, rather strong	Colorless Pale yellow	0.92	30.7	0.14	0.9	None	1: 34.1
192	Good sandy soil (4)	91.4	None		4	Neutral	Neutral	Slightly alkaline	Colorless Pale yellow	0.94	31.3	0.25	1.7	None	1: 18.4	
382	Clay soil, poor in humus (8)	86.2	None	0	Neutral—slightly alkaline	Neutral	Neutral	Slightly alkaline	Colorless Pale yellow	0.78	26.0	0.15	1.1	None	1: 23.6	
881	Good sandy soil, rather rich in humus (5)	94.2	Very slight	1	Neutral—slightly alkaline	Neutral	Slightly alkaline	Slight	Colorless Pale yellow	1.19	39.7	0.05	0.3	None	1: 132.3	
079	Loam soil, poor in humus (5)	91.8	None	4	Neutral—slightly alkaline	Neutral	Neutral—slightly alkaline	Neutral	Pale yellow	0.78	26.0	0.10	0.7	None	1: 37.1	
989	Good sandy soil, rich in humus (6)	92.0	None	4	Neutral—slightly alkaline	Neutral	slightly alkaline	Alkaline	Colorless Pale yellow	0.57	19.0	0.05	0.3	None	1: 63.3	
2233	Good sandy soil (4)	88.0	Slight	4	Neutral—slightly alkaline	Neutral	slightly alkaline	Alkaline	Colorless Yellow	0.86	28.7	0.12	0.8	None	1: 35.9	
997	Sandy loam soil (5)	91.8	Very slight	4	Slightly alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.48	16.0	0.01	(-0.1)	None		
065	Sandy loam soil, poor in humus (4)	91.6	Slight	4	Slightly alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.25	8.3	-0.09	(-0.6)	None		
011	Dark sandy soil, rather rich in humus (5)	86.0	Very slight	3	Slightly alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.63	21.0	--**	None			
238	Good sandy soil, rich in humus (7)	84.2	Very slight	4	Slightly alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.54	18.0	±0.00	±0.0	None		
237	Good sandy soil, rich in humus (7)	87.4	Strong	4	Slightly alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.64	21.3	0.03	0.2	None	1: 106.5	
001	Sandy loam soil, poor in humus (4)	96.0	Very slight	4	Alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.38	12.7	-0.05	(-0.3)	None		
179	Loam soil (6)	90.4	Very slight	4	Alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.38	12.7	±0.00	(±0.0)	None		
142	Loam soil, rather rich in humus (6)	85.6	Slight	4	Alkaline	Alkaline	Alkaline	None	Colorless Yellow	0.24	8.0	0.05	0.4	None	1: 20.0	

TABLE II (Cont.)

SOIL SAMPLE NUMBER	PER CENT DRY MATTER IN THE SOIL	ACID EFFERVESCENCE	AMMONIUM VIBRATION	PER CENT AMMONIUM CHLORIDE-SOIL	REACTION FOR LUMINESCENCE SOLUTION	REACTION FOR LUMINESCENCE PAPER (SOIL RING)	REACTION FOR LUMINESCENCE PAPER OF SOIL MIXTURE	REACTION FOR POTASSIUM IODATE (BAUMANN AND GULLY'S METHOD)	ACID TESTED, EXPRESSED IN CUBIC CENTIMETERS 1/10 N ACID	REACTION FOR POTASSIUM IODATE AND POTASSIUM CHLORIDE METHOD		Potassium chloride method	Acetate method	The absolute acidity of per 100 grams soil dry soil per 100 grams dry soil	Soil moisture per 100 grams air-dry soil	Soil moisture per 100 grams air-dry soil	Color of the starch solution	Color of the liquid filtrate	TESTS OF THE GELATION REACTIONS WITH SODIUM ACETATE AND WITH THE POTASSIUM CALCIUM SULFATE			
										Reaction for Potassium iodate	Reaction for Potassium chloride								TESTS OF THE GELATION REACTIONS WITH SODIUM ACETATE AND WITH THE POTASSIUM CALCIUM SULFATE			
4014	Good sandy soil (4)	89.6	Very slight	4	Alkaline	Alkaline	Alkaline	Alkaline	None	Colorless	Yellow	0.18	6.0	-0.10	(-0.7)	None						
4018	Sandy loam soil (5)	87.2	Very slight	4	Alkaline	Alkaline	Alkaline	Alkaline	None	Colorless	Yellow	0.22	7.3	-0.10	(-0.7)	None						
4167	Clay soil, rather rich in humus (9)	83.6	Strong	4	Alkaline	Alkaline	Alkaline	Alkaline	None	Colorless	Yellow	0.26	8.7	-0.05	(-0.4)	None						
4276	Good sandy soil (4)	95.2	Very strong	4	Strong alkaline	Strong alkaline	Alkaline	Alkaline	None	Colorless	Yellow	0.12	4.0	-0.05	(-0.3)	None						
4245	Sandy loam soil (5)	91.2	Very strong	4	Strong alkaline	Strong alkaline	Alkaline	Alkaline	None	Colorless	Yellow	0.22	7.3	-0.05	(-0.3)	None						
1. Field Soils																						
1	Raw high-bog peat from "Knundermose" near Herning	85.7	None	0.014	Strong acid	Strong acid	Strong acid	Strong acid	Acid	Acid	Acid	Very strong	Light brown	Blue	Colorless	1.90	1140	3.92	235	Quite large		
2	Raw high-bog peat from "Vejen Mose"	87.9	None	0.021	Strong acid	Strong acid	Strong acid	Strong acid	Acid	Acid	Acid	Very strong	Dark brown	Blue	Colorless	2.12	1272	5.12	307	Large 1: 4.1		
3	Raw high-bog peat from "St. Vildmose"	84.4	None	0.061	Strong acid	Strong acid	Strong acid	Strong acid	Acid	Acid	Acid	Very strong	Medium brown	Blue	Colorless	2.16	1296	3.33	200	Quite large 1: 6.5		
2. Forest Soils†																						

				Neutral	Neutral—slightly alkaline	Slightly alkaline	None	Colorless	Pale yellow	0.64	384	0.37	22	None	1: 17.5	
4	Peat from the transition belt between high-bog and low-bog in "St. Vildnose," Gjelleruplund near Herning	82.4	None	42.16	Neutral	Neutral—slightly alkaline	Strongly alkaline	Colorless	Pale yellow	0.64	384	0.37	22	None	1: 17.5	
5	Low bog peat from Brosgaard near Vejen	56.7	None	0.146	Acid	Acid	Slightly acid	Yellow	Blue	Colorless	1.50	900	0.87	52	None	1: 17.9
6	Low-bog peat from Tylstrup	87.9	None	0.219	Slight acid	Neutral—slightly acid	Acid	Strong	Pale blue	Colorless	1.02	612	0.37	22	None	1: 27.8
7		74.2	None	0.139	Acid	Acid	Acid	Very pale yellow	Blue	Colorless	1.86	1126	1.07	64	None	1: 17.6

* In the case of peat soils: per 1/6 gram oven-dry soil.

† In the case of peat soils: per 100 grams oven-dry soil.

‡ In the case of peat soils: per 5 grams oven-dry soil (= half of the liquid).

** There was too little soil to make a quantitative determination. A specimen with a smaller amount of soil (14 grams in 70 cc. of the liquid) showed, however, that the potassium chloride extract gave a distinct alkaline reaction for phenolphthalein.

†† All the soil samples come from the upper 30 cm. peat layer.

To illustrate the matter further, observations were made of the size of the gelatinous precipitate appearing when potassium chloride extract is saturated with sodium hydroxide. In connection with these investigations, with the help of a strip of red and of blue litmus paper placed over the surface of the solution before filtering, and left for about an hour, observations were made of the reaction of the potassium chloride solution after it had stood five days with the soil. The results of the investigations are given in table 11.

As litmus is the indicator which has hitherto been used in qualitative investigations of the reaction of the soil made by the author, the results obtained from them are used as a basis for comparison. As was the case in earlier investigations [see further (10, p. 431)] the reactions from the litmus solution are expressed in the following way:

Strongly acid (yellowish-red color).

Acid.

Slightly acid.

Neutral—slightly acid.

Neutral (violet color).

Neutral—slightly alkaline.

Slightly alkaline.

Alkaline.

Strongly alkaline (strong blue).

Litmus-test. As is stated before, investigations have been made with the use of litmus solution and litmus paper. With the former we have also made an investigation comparing the reaction of the soil specimens when in the moist condition in which they were at the beginning of the experiment, and in an air-dry condition. As table 11 shows, the drying has had no effect upon the reaction which may be positively stated—in any case only shades of distinction (to the one side or to the other). Therefore, in making qualitative reaction-determinations it seems to be a matter of indifference whether the soil is used in a moist or an air-dry condition, and the somewhat larger amount of CO₂ which we take for granted is present in moist soil, has no appreciable influence on the determinations.

In many soils, and especially in the loam and clay soils, the reactions in the litmus solutions appear sharpest and most distinct when dry soil is used, for the liquid becomes more quickly and easily clear.

The presence of a larger amount of fine particles floating about gives the litmus solution a more reddish color than the reaction itself causes, and on this account the determination is somewhat uncertain. In such cases the litmus-paper test is to be preferred. On the whole the results obtained from this method correspond satisfactorily with those gained by using the litmus solution; but the latter is generally better able to express small differences in the reaction of the soil. As the strongly acid reaction (the characteristic yellowish-red color in the litmus solution), does not appear at all in the paper-test, the litmus solution must be considered to give more complete

information about the reaction of the soil than litmus paper, and as the former method is quicker, which is of no small importance when examinations are to be made on so large a scale as the case is here in Denmark,¹⁵ that method is on the whole to be preferred. In cases where a doubt arises (very opaque liquids, see above), the litmus paper test should always be used for control, and as it does not require much practice for making a correct valuation of the color changes, it would perhaps also be the best method to use when the determinations are made by persons of little experience.

Daikuhara's nitrite-method. As the table shows, this method is very sensitive to small changes, all the soils which in the litmus-test have been proved more or less acid (strongly acid—slightly acid), have caused a strong blue color to appear on the potassium iodate starch paper. The same is true of various neutral soils both with and without *Azotobacter* vegetation. First, when litmus shows a decidedly alkaline reaction (slightly alkaline—strongly alkaline), the blue color ceases to appear on the paper. Therefore, according to the investigations previously made of the lime-requirement of the soil (10), we find a large number of non-lime-requiring soils which tested by Daikuhara's method give an acid reaction. On the other hand, it is fairly safe to say that all the soils which do not color potassium iodate starch paper blue have no lime-requirement. Yet, as all these soils, either by the litmus test alone, or in connection with the *Azotobacter* test prove to be rather rich in basic substances, the nitrite method does not fill any particular need, either in making a general determination of the reaction of the soil or of its lime-requirement. Yet to have demonstrated the ease and speed with which this reaction takes place, is of interest in another connection, for it suggests that to use nitrites as nitrogen fertilizers, which has often been under consideration, would for many types of soil risk a considerable nitrogen loss.

The iodine method according to Baumann and Gully. The results of the determinations based on the Baumann-Gully method on the whole agree well with the results of the reaction-determinations made by the litmus-test. All the soils which have shown a distinct acid reaction for litmus have caused a distinct yellow coloring of the solution used. In the case of the neutral soils the liquid is either slightly yellow or colorless, and no distinction can be made between neutral soils with and without *Azotobacter* vegetation. In using soils that show a positive alkaline reaction for litmus solution (slightly alkaline—strongly alkaline), no iodine has been freed.

Nor does this method give information as to the reaction of the soil other than has appeared from the litmus test, and as the method is rather detailed, and the reagents very sensitive to the CO₂ in the air, we need not expect that it will play an important part in making a qualitative determination of the reaction of the soil.

P-nitrophenol method. The results of the reaction-determinations when this

¹⁵ In the years 1910-1916, using the combined litmus and *Azotobacter* test (1 to 1) suggested by the author, about 40,000 soil specimens were examined.

indicator is used agree in general very well with those with the litmus solution. The soils which show acid reaction for litmus have either—and this is usually the case—decolored the solution, or else caused no change in the original very slightly yellowish tinge of the liquid. In the case of soils showing a neutral reaction for litmus, there is a distinct difference between the two groups, with and without *Azotobacter* vegetation, for the former colors the liquid somewhat more yellow than the latter. There are, however, a few exceptions to this rule, the soils which for litmus are decidedly alkaline show, with one or two exceptions, among the slightly alkaline soils, the very strongest possible yellow color (greenish-yellow). The use of p-nitrophenol must on the whole be considered to give very good information as to the reaction of the soil, and in certain cases where the litmus test has shown a neutral reaction, this indicator may be used with advantage in supplementary investigations.

However, litmus has this one advantage over all other indicators—it shows distinct changes on both sides of the neutral point, and on this account no other indicator can supplant it in ordinary qualitative determinations of the reaction of the soil.

In addition to these named, experiments have been made with various other indicators—for example "neutral red," which like litmus has its turning-point in the neighborhood of the true neutral point. Unfortunately, this indicator can not be used in reaction-determinations of the soil, as it is to a larger or smaller extent discolored by it.

In connection with these investigations, an experiment has been made with a colorimetric measurement of the hydrogen-ion concentration in the filtrates from the mixtures of field soils and water. The experiment has given negative results in that there seems to be an indication that the colorimetric method cannot generally be used in determining hydrogen-ion concentration in the watery soil-extracts, a condition which must be largely explained by an insufficient appearance of "buffers" [see further, S. P. L. Sørensen (3, p. 17)]. There is a large quantity of these present in the soil itself. In a colorimetric determination of the reaction of the soil, we are reduced to working with mixtures of soil and water, and an exact determination of the hydrogen-ion concentration in watery soil-extracts can, therefore, in general be made only by the electro-metrical method.

In connection with the investigations of the methods for a qualitative determination of the reaction of the soil referred to above, quantitative determinations of the ability of the soil to free acid have been made, the potassium chloride method suggested by Daikuhara being used in part, and the calcium acetate method in part.

The main results of the investigations of mineral soils are grouped together in the summary in table 12, and in figure 4, which deals particularly with the soils with which both the calcium acetate and the potassium chloride method have been used.

As this summary shows, the ability of the soil to free acid, measured ac-

cording to both methods, is, on the average, greatest in those soils giving an acid reaction for litmus, and decreases with decreasing acidity and increasing alkalinity (for litmus). But aside from this agreement in the principal characteristic, the two methods show very important and characteristic differences in the results of the investigations.

First, in considering the relation of the soils to the potassium chloride solution we find that *only those soils showing a decidedly acid reaction for litmus are able to free acid from this solution to an appreciable extent*; and it is important to note (table 11), that this ability, though varying greatly in degree, is yet present in all soils showing an acid reaction.¹⁴ The amount of acid freed from the potassium chloride solution decreases greatly as the neutral reaction

TABLE 12
Relation between the reaction of mineral soils for litmus and their ability to free acid

REACTION FOR LITMUS	NUMBER OF SOILS	ACID FREED, EXPRESSED IN CUBIC CENTIMETERS OF ACID 1/10 N PER 100 GRAMS AIR-DRY SOIL								RELATION BETWEEN THE AMOUNT OF ACID FREED BY THE POTASSIUM CHLORIDE METHOD AND THAT FREED BY THE CALCIUM ACETATE METHOD	
		Acetate method				Potassium chloride method					
		Largest amount of acid freed	Smallest amount of acid freed	Difference between the largest and smallest amounts of acid freed	Average	Largest amount of acid freed	Smallest amount of acid freed	Difference between the largest and smallest amounts of acid freed	Average		
Strongly acid.....	13	150.0	46.0	104.0	82.9	150.3	5.7	144.6	35.6	1: 2.3	
Acid.....	10	100.7	43.3	57.4	66.8	43.3	2.3	41.0	10.3	1: 6.5	
Slightly acid.....	9	56.7	30.7	26.0	48.2	10.4	1.1	9.3	3.8	1: 13	
Neutral—slightly acid.....	9	63.3	22.3	41.0	43.5	5.3	0.6	4.7	1.9	1: 23	
Neutral.....	18	70.3	17.3	53.0	33.8	1.7	0.3	1.4	0.8	1: 42	
Neutral—slightly alkaline.....	4	39.7	19.0	20.7	27.7	1.1	0.3	0.8	0.6	1: 46	
Slightly alkaline.....	6	28.7	8.3	20.4	16.9	0.8	-0.6	1.4	0		
Alkaline and strongly alkaline.	8	12.7	4.0	8.7	8.3	0.4	-0.7	1.1	-0.3		

is approached, and at that point the amount of acid freed is so small that in the case of field soils a quantitative determination is inaccurate. (It must be remembered that the figures expressing the absolute acidity are obtained by multiplying the values found with the factor 3.) *The variations in the ability of this group of soils to free acid as well as in the following groups are very small.*

¹⁴ This close agreement between the results of the potassium chloride and the litmus methods may be said to make even more probable the supposition (page 144) that the ability to color litmus red (in opposition to Baumann and Gully's opinion) is caused by the presence of *truly acid-reacting substances* in the soil in question.

G. Fischer reaches a similar result by another method. By an electro-metric measurement of the hydrogen-ion concentration in various peat soils he proves that in those showing an acid-reaction for litmus there is a considerable hydrogen-ion content.

The investigations of peat soil made (table 11, show that high-bog peat specimens have a far greater ability to free acid from potassium chloride than low-bog peat specimens, even though the latter show an acid reaction for litmus. When an acetate solution is used, the difference between the amount of acid freed by high-bog peat and that freed by low-bog peat is far less apparent, and one of the low-bog peat specimens (no. 7) even shows almost as great an amount of acid freed as the high-bog peat specimens.

The slightly blue color in the litmus solution (slightly alkaline reaction—a reaction almost coincident with the absolute neutral point, which is charac-

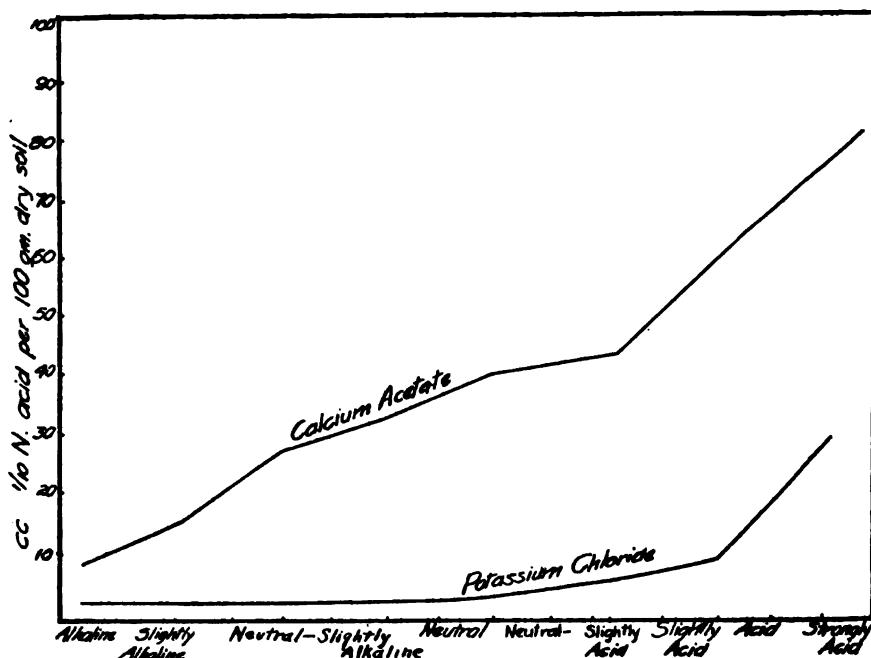


FIG. 4. DIAGRAM SHOWING THE RELATION BETWEEN THE REACTION OF THE SOIL FOR LITMUS AND ITS ABILITY TO FREE ACID

terized by the presence of an equal number of H and OH-ions) indicates that acid is no longer being freed from the potassium chloride solution, and where the reaction is more alkaline (for litmus), the potassium chloride solution is in almost every instance distinctly alkaline (for phenolphthalein).

The results at hand may, with considerable certainty, be said to suggest that the freeing of acid from the potassium chloride solution is alone determined by the presence of *truly acid-reacting substances* in the soil in question. This condition was already apparent in the results, shown in table 11, of the qualitative investigations of the reaction of the potassium chloride solution, which, after standing for five days with the soil, was tested with litmus paper.

These results agree very well with those from reaction-determinations made according to the normal litmus method.

As has been mentioned before, Daikuhara states that when a surplus of ammonium is added to the filtrate of the potassium chloride solution, which has stood with acid mineral soil, a gelatinous precipitate always appears. This is composed of aluminum—or (in certain instances), iron-hydroxide, and the proportion always bears a definite relation to the acidity of the solution. Therefore, he concludes that the acidity of mineral soils is solely dependent on the appearance of certain acid aluminum—or iron—compounds (acid silicates).

To throw further light on the question notes have been made of the size of the precipitate appearing when the potassium chloride solution is saturated with sodium hydroxide. The results appear in table 11.

As may be seen, *all strongly acid; and, moreover, the majority of acid or slightly acid, mineral soils, give a whitish gelatinous precipitate, whereas no investigation has ever shown this to be true of neutral or slightly alkaline soils.* The amount of the gelatinous precipitate increases with the increased acidity of the liquid (measured according to the potassium chloride method), and when the number expressing this is over 6, the precipitation has always taken place.

These observations seem to confirm Daikuhara's statements, even though they are unable to give positive assurance as to whether the acidity of the mineral soils depends, as that author claims, solely on the presence of substances of the above-mentioned kind.

In humus soils we notice that in neutralizing the extracts from the high-bog peat samples, a considerable gelatinous precipitate appears (in this instance reddish-brown in color), and that the amount of the precipitate (as in mineral soils) seems to bear a certain relation to the amount of acid freed from the potassium chloride solution. One would thus be inclined to suppose that the acidity of this type of soils depends to a large extent on acid iron compounds. Special research work on this point would have particular interest.

Low-bog peat specimens, which, as has been said, have a somewhat limited power to free acid from a potassium chloride solution, showed, after the potassium chloride extract had been neutralized, no gelatinous precipitation, even though the soil specimens themselves gave a decidedly acid reaction for litmus.

In addition to the determinations already mentioned, a determination has been made according to methods previously used (10), of the lime in peat soils which is soluble in ammonium chloride. As table 11 shows in the two humus types examined, no connection has been found between the acid freed from acetate or potassium chloride solution, and the content of lime soluble in ammonium chloride in the soil.

If we now examine the results from the acetate method, we will immediately see that these are far greater than those obtained from the potassium

chloride method. This is due to the fact either that in using the latter method one is unable to obtain as correct an expression for the absolute acidity of the soil as from the acetate method, or that the acid may be freed from the calcium acetate solution not only by the action of *truly acid-reacting substances*, but also by substances of another character.

In table 12 is a summary of the results of the investigations of mineral soils. From these statistics it appears that the values expressing the degree of acid obtained according to the acetate method, in contrast to those from the potassium chloride method, diminish regularly from acid to alkaline reaction, and that thus both neutral and alkaline soils have the power of freeing a considerable amount of acid from an acetate solution. Resulting from this the relation between the amounts of acid freed from potassium chloride and calcium acetate is far closer in distinctly acid than in approximately neutral soils.

This condition appears plainly in figure 4, showing the relation between the reaction of mineral soils for litmus, and the degree of acid freed from the two salt solutions named. While the curve representing the absorption of bases in the potassium chloride solution runs almost parallel with the abscissa axis until acid reaction begins and then rises quite sharply, the curve representing the calcium acetate solution never reaches the abscissa axis, and rises gradually. A closer examination of the single determinations made, calcium acetate solution being used, table 11, shows that the relation between the acid freed and the reaction of the soil is far from being as regular as this diagram would seem to indicate. While the variation in the ability of neutral soils to free acid from potassium chloride solution is always very small, and the amount of acid freed is always much less than in distinctly acid soils, when calcium acetate solution is used with neutral soils the variations in the amount of acid freed are very large, and neutral field soils quite frequently free even a larger amount of acid than very distinctly acid-reacting soils.

These characteristic differences in the results obtained from the two methods are presumably due to the fact that the absorption of bases, and the consequent freeing of acid from the calcium acetate solution is not, as is the case with potassium chloride solution, solely caused by the presence of *truly acid-reacting substances*, but also by the presence of substances of quite another character, whether these be adsorptively unsaturated colloids, or other non-acid reacting substances with the ability to absorb bases from salts of weak acids.

In peat soils it is interesting to note (table 11) that in the relation between the two amounts of acid freed from the two liquids there is a characteristic difference between high-bog peat and low-bog peat, the relation being 4 to 5 times closer in high-bog peat than in low-bog peat.

As has been mentioned before (page 118), Daikuhara claims that by using his potassium chloride method one is able to express quantitatively the lime-

requirement of the soil. If his claim is correct, only those soils giving acid reaction for litmus (based on the investigations mentioned above) have a distinct lime-requirement while all neutral soils either have no lime-requirement whatsoever or a very small one.¹⁷ Previous investigations made by the author and O. H. Larsen (10) show that a large number of field soils with neutral reaction for litmus have a decided lime-requirement, and that when lime is added to these soils, sometimes a greater result is obtained than from the distinctly acid soils. In such instances, adding the 100 kgm. of carbonate of lime, which according to the results obtained from thi method would usually suffice, is practically without effect. And too, with many of the distinctly acid soils, measuring the amount of lime according to the results of the potassium chloride method will give far too small an amount for practical purposes. For many decidedly acid soils, the figures expressing the absolute acidity are between 5 and 10, and in such cases, according to the method of computing given here, the addition of 600 to 1200 kgm. of carbonate of lime per hectare, should be sufficient to fulfill the lime-requirement in the soil. However (partly due to the fact that in the field it is impossible to mix the lime with the acid soil so thoroughly as in the retorts in the laboratory), and judging from practical experience and the results of the field experiments, such small amounts of lime are far too insignificant entirely to fulfill the lime-requirement of the soil.

However, even though by determining the acidity of the soil we are unable to state positively the extent of its lime-requirement, yet a quantitative acid determination can in many cases give suggestions as to the magnitude of this need, and there is no doubt that soil specimen no. 3313, showing an acidity content corresponding to *ca.* 150 cc. of 1/10 N acid per 100 grams of dry soil requires a considerable amount of lime for satisfying its lime-requirement, for according to this method of computing, it is necessary to add *ca.* 18,000 kgm. of carbonate of lime per hectare merely to neutralize the soil.

Of the methods which at the present time seem to give the best information in regard to the degree of the acidity of the soil, the potassium chloride method is the one most deserving of attention, and its use will in many instances be a great assistance in making a diagnosis of the condition of the soil.

It is generally supposed that a larger amount of lime is necessary for satisfying the lime-requirement of clay soils than of sandy soils. If this supposition is correct, we have reason to expect that the former soils within the single reaction groups (and the groups with and without *Azotobacter* vegetation) possess a greater ability to absorb bases than the latter.

To throw light on the matter the material at hand was treated as table 13 shows. This summary includes the results obtained from table 11, and in the

¹⁷ An amount of acid freed corresponding to 1 cc. 1/10 N acid for 100 grams soil will only require *ca.* 120 kgm. CaCO₃ for neutralizing the acid content in the upper 20 cm. per hectare (see further, page 145).

case of the acetate method those from table 6 as well. A distinction is made among 5 degrees of heaviness in soil, which in the last division of the table are grouped together in two classes—clay soil and sandy soil.

In looking over the results obtained from the acetate method, we see that the clay soil in all three reaction groups and in the groups with and without *Azotobacter* vegetation possesses, on the average, somewhat less ability to absorb lime than the sandy soils of the corresponding groups. In using the potassium chloride method the relation is quite the opposite, for the clay soils with acid reaction—or by the *Azotobacter* test—without the ability to cause *Azotobacter* development, possess on the average more than twice as great

TABLE 13
Comparison between the ability of clay and of sandy soils to free acids

CHARACTER OF THE SOIL	ACETATE METHOD								POTASSIUM CHLORIDE METHOD											
	Number of soils			Acid freed in cm. [‡] 1/10 N acid per 100 gm. dry soil					Number of soils			Acid freed in cm. [‡] 1/10 N acid per 100 gm. dry soil								
	Acid*	Neutral	Alkaline†	Without az. v. §	Acid	Neutral	Alkaline	Without az. v.	Acid	Neutral	Alkaline	Without az. v.	With az. v.							
		With az. v.	With az. v.	With az. v.		With az. v.	With az. v.	With az. v.		With az. v.	With az. v.	With az. v.	With az. v.	With az. v.	With az. v.					
Clay soil.....	9	8	6	13	10	62.7	37.8	14.2	55.0	23.7	7	6	1	11	3	37.2	1.1	-0.4	24.2	0.3
Loam soil.....	7	4	3	8	6	65.2	33.4	17.5	60.6	26.4	5	3	2	6	4	20.6	0.6	0.0	17.3	0.3
Sandy loam.....	4	8	7	6	13	47.8	30.0	9.8	43.0	18.6	3	7	6	5	11	9.8	0.8	-0.3	6.2	0.2
Good sandy soil.....	11	13	8	16	16	82.1	41.0	15.9	72.7	25.0	10	11	5	15	11	12.6	0.9	-0.1	8.7	0.4
Light sandy soil.....	11	11	1	21	2	67.8	37.5	21.1	53.1	32.5	7	4	0	11	0	11.5	2.6	—	8.3	—
Loam soil.....	20	20	16	27	29	60.6	33.8	12.9	54.0	22.0	15	16	9	22	18	26.2	0.9	-0.2	18.2	0.3
Sandy soil.....	22	24	9	37	18	75.0	39.4	16.5	61.5	25.8	17	15	5	26	11	12.1	1.3	-0.1	8.5	0.4

* Includes groups: Strongly acid, acid and slightly acid.

† Includes groups: Neutral-slightly acid, neutral and neutral.

‡ Includes groups: Slightly alkaline, alkaline and—slightly alkaline,—strongly alkaline.

§ az. v.—*Azotobacter* vegetation.

an ability to free acids as the sandy soils of these groups. In neutral and alkaline soils, and in those soils causing *Azotobacter* development, the ability to free acid from potassium chloride is, as has been mentioned before, so small that it is impossible to indicate the differences in the degree of this ability for various soils.

As the comparison made shows, a different result is obtained according to whether the calcium acetate or the potassium chloride method is used. It is impossible for the present to state which of these two methods can on the whole give the most helpful information as to the lime-requirement of the soil. With these data we are unable to verify the extent to which the above-named supposition that lime-poor clay soils require on the average a greater addition of lime than sandy soils is correct. It is indeed probable that the

supposition is correct in the case of mineral soils showing an acid reaction for litmus, for the results appearing from the potassium chloride method suggest decidedly that acid clay soils require on the average a far greater amount of basic substances to attain a neutral reaction than acid sandy soils.

III. SUMMARY OF THE MAIN RESULTS OF THE INVESTIGATIONS

As a result of the investigations made, it seems to be necessary to distinguish sharply between *the absolute acidity of the soil*, and *its ability to free acids* (absorb bases).

The majority of methods suggested for making a quantitative determination of the acidity of the soil give no information as to the content of *truly acid-reacting substances* in the soil, *i.e.*, substances which in solution cause a hydrogen-ion concentration greater than corresponds to the value 10^{-7} ; but only of its ability to absorb bases, which is partly determined by the presence of acid-reacting substances, and partly by the presence of non-base saturate colloids, or other non-acid reacting, but base-absorbing substances.

We still lack a method for making a positive quantitative determination of the acidity of the soil, in the meaning of the word given above, even though the methods suggested by Hopkins and his collaborators, and later by Daikuhara with the use of chloride solutions, seem in many cases to give valuable information on this point.

In determining the ability of the soil to absorb bases, a method based on Baumann and Gully's principles, in which acetates are employed, should be preferred to Tacke and Sütching's method, not only because it gives results which are more certain and approach the absolute values more closely, but also because the investigations may be made far more quickly than according to the latter method.

Baumann and Gully's claim that no free acids exist in sphagnum peat is probably incorrect. The results of the investigations made here must be understood to mean that the ability to absorb bases characteristic of this type of soil, as well as of others which show an acid reaction for litmus, is in every instance partly dependent on the appearance of *truly acid-reacting substances*, a result which coincides fairly well with the claim made in a former paper (9), that raw high-bog peat contains a considerable amount of such substances. The further claim made by the above-mentioned scientists, and later repeated by Ramann, that the ability of a soil to color a neutral litmus solution red need not be an expression for the presence of free acids in it, but is solely caused by the ability of soil colloids to free acids from neutral salts, is also incorrect, for it has been proved that there is no closer connection between the ability of the soil to free acids, determined according to the calcium acetate method, and its reaction for litmus, and furthermore, that many of the soils which when tested by this method show a considerable ability to free acids, show a neutral or slightly alkaline reaction for litmus. The fact

that it is only those soils with a distinct acid reaction for litmus which are able to an appreciable extent to free acid in a potassium chloride solution, must, on the contrary, be said to indicate that the ability of the soil to color neutral litmus solution red is due to *the presence of truly acid-reacting substances* in the soil in question. The importance of the litmus test in making soil investigations, especially in examining the requirement of the soil for basic substances, which in practice is its lime-requirement, is therefore easily understood.

A determination of the base-absorption power of the soil is not sufficient for determining its lime-requirement, for it has been found that many of the soils, which, based on former investigations, were considered to require lime (such as those not causing *Azotobacter* vegetation under the *Azotobacter* test) possess less power to free acids in a calcium acetate solution than those which do not require lime (such as those causing *Azotobacter* vegetation under the *Azotobacter* test). Nor can we expect a determination of the acidity of the soil to give positive information as to whether or not it requires lime. As has been proved previously, all mineral soils showing an acid relation for litmus have a large lime-requirement. In such instances a quantitative determination of the acid content of the soil which expresses the amount of lime necessary for neutralizing the acid present, will also give reliable though insufficient information as to the degree of the lime-requirement of the soil. However, we must remember that many soils which show a neutral reaction for litmus, have a decided lime-requirement and in such cases it is impossible to use the acidity determination in determining the degree of the lime-requirement of the soil.

At this point it is well to emphasize the fact that the qualitative determination of the lime-requirement, according to the combined litmus and *Azotobacter* tests in general use in Denmark at present, can to a certain extent give information as to the degree of the lime-requirement of the soil. Acid-reacting soils which only to become neutral require a certain amount of lime, doubtless need, on the average, a far more considerable lime addition to fulfill their lime-requirement than neutral lime-requiring soils without *Azotobacter* vegetation, which presumably in the majority of instances should be and are content with the addition of a proportionately small amount of lime. To elucidate further the question which has great practical importance, investigations must be made in connection with a larger number of field experiments in the use of lime.

As the author has previously stated, it is probable that the question of the lime-requirement of the soil is predominantly a question of *the presence or absence of certain easily-decomposed acid-saturating calcium (or magnesium) compounds*. If the problem is such, the efforts to find a quantitative expression for the lime-requirement must be in the direction of methods which can express the amount of lime-fertilizer necessary for supplying the soil with a

sufficient quantity of such compounds. To what extent there are possibilities for finding such a method, and whether or not it should rest upon a chemical or a biological foundation, must for the present remain unanswered.¹⁸

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¹⁸ Since the above article appeared, H. Kappen has published an interesting investigation of the reaction of the soil (*Studien an sauren Mineralböden aus der Nähe von Jena*.—In *Landw. Vers. Stat.*, Bd. 88, 1916, p. 13), of which, unfortunately, no account could be taken.—H. R. C.

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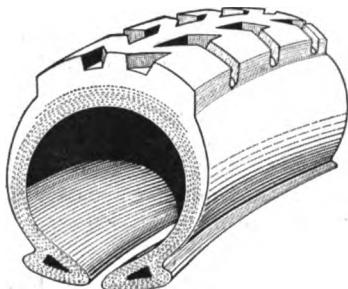
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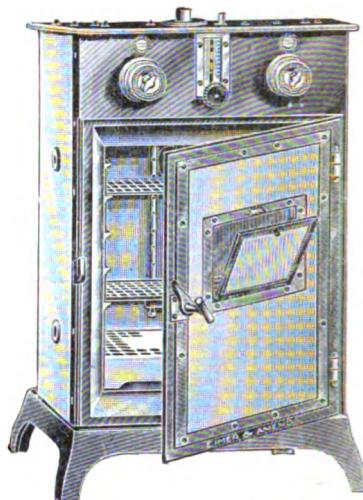
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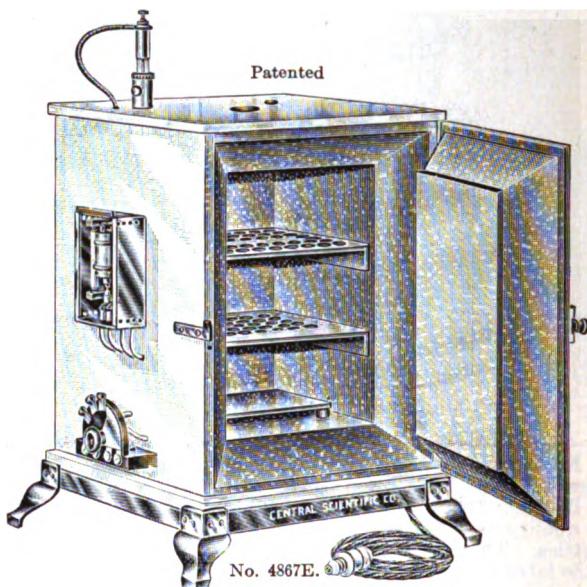
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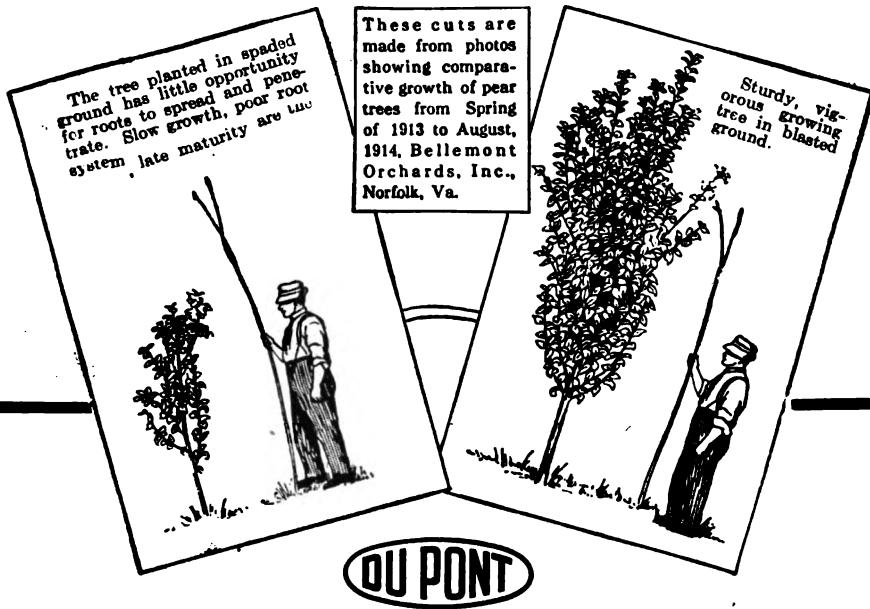
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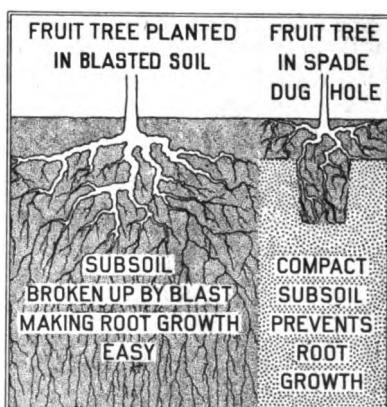
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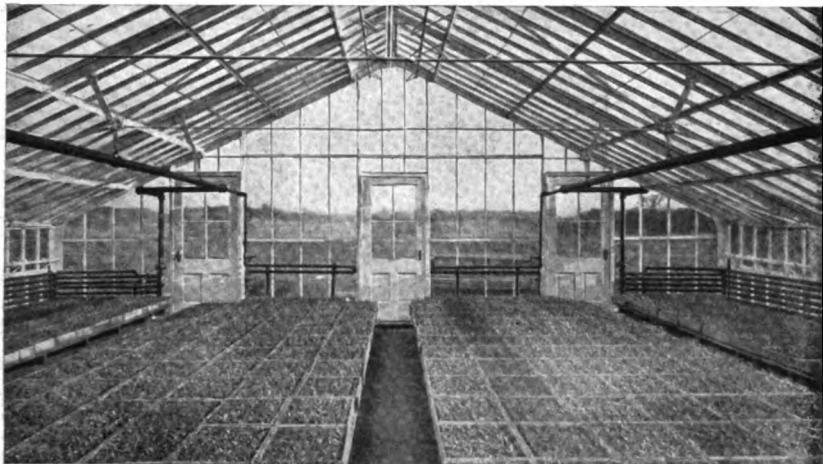
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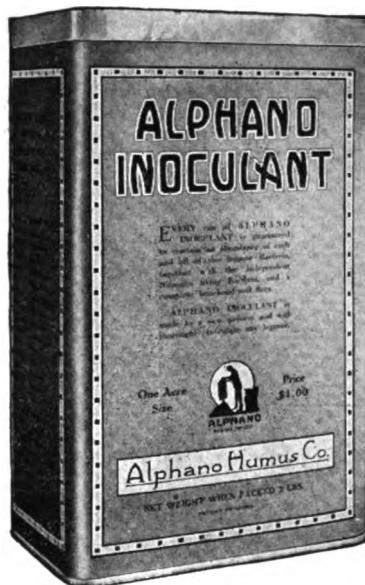
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SOME FACTORS INFLUENCING THE QUANTITATIVE DETERMINATION OF NITRIC NITROGEN IN THE SOIL

J. E. GREAVES AND C. T. HIRST

From the Utah Agricultural Experiment Station

Received for publication April 10, 1917

For a long time man has been intensely interested in studying the nitric nitrogen of the soil. He has learned that its presence in a soil is necessary for the production of plants, and hence is essential to his own existence. The nitric nitrogen in water is used as a measure of its purity. Recently it has been used in the manufacture of many synthetic compounds; especially is this true of explosives. Civilized nations have therefore come to prize greatly those substances which are rich, or comparatively rich, in nitric nitrogen. Hence, the literature on the subject is voluminous, and many methods have been developed for the determination of nitric nitrogen. Some of these are accurate and workable if used on the substance for which they were intended, and is used under suitable conditions. Others scarcely reach the dignity of qualitative tests. The more important of the methods have been listed and described by others (56, 18). Hence, it is needless for us to consider them in detail here. The majority of the methods have been developed for the determination of nitric nitrogen in water, fertilizers, and various substances comparatively easy to handle. Many are known to be inaccurate when used on substances rich in chlorine, while others are unreliable if used on material high in organic matter. The conditions requisite for the successful use of some of the methods when used on substances high in soluble salts or various colloidal materials are not fully understood; hence, this work has been undertaken with the hope of perfecting a method suitable for the determination of nitric nitrogen in soils, especially those rich in soluble salts.

A method for the determination of nitric nitrogen in soil must be accurate when applied to substances containing either much or little nitric nitrogen. It must be accurate when used on soils having large quantities of soluble salts and those containing large amounts of organic matter. It should be fairly easy of manipulation so that a great number of determinations can be made in a short time.

This last factor would eliminate all of the gasometric methods, for, while accurate, they require considerable time as well as expensive complicated apparatus. The Busch nitron (11) method is not applicable to soil as there are many substances in the soil which interfere (31, 39) in the method, and moreover, the reagent is very expensive. Organic matter interferes in the

Devarda (14), Tiemann-Schulze (22), Baussingault indigo (23) and Schlösing (38) methods, while organic matter is claimed to be without effect on the Ulsch (41) method.

Results obtained by the electrolytic reduction method vary with a number of factors, namely, the concentration (25) of the acid, strength of current, kind of cathode (36), the deposition of copper at the cathode, whether smooth, spongy, or amalgamated, while chlorides (53) if present interfere.

The sodium amalgam method yields a mixture of products (15) according to the conditions of the experiment. Among these are nitrogen, nitrous oxide, hydroxylamine, hyponitrite and ammonia. The tip reduction method is not reliable in the presence of organic matter, and the reduction to hydroxylamine is not quantitative (28). Furthermore, neither it (32) nor the phenylamine (16) method is applicable in the presence of ferric salts.

The titration of nitrates with ferrous sulfate (5) is not suitable for the determination of small quantities, while chlorides interfere. The results of the brucine method (29) vary with the acid and are not quantitative. The phenoldisulphonic acid method, while probably one of the most accurate for the determination of nitric nitrogen, is not applicable to colored solutions unless first clarified and the organic matter oxidized (49) and soil salts—especially the chlorides and sulfates—cause a loss of nitrates (30, 45).

The iodometric (19, 54) method is unsuited for the determination of nitrates in soil, as many factors interfere and the distillation must be conducted in a current of carbon dioxide. Even under this condition it is far from being as accurate as many of the other methods.

Alberti and Hempel (1) obtained excellent results with the Ulsch method on both pure nitrates and complex manures. L. von Wissel (54) used the Devarda and the Ulsch methods on solutions containing 16.5 per cent of nitrates. With the Ulsch method he recovered 16.38 per cent, while with that of Devarda he obtained 16.42 per cent. Schmöger (43) in comparative tests with the Raab-Böttcher, Ulsch, Devarda, and Kjeldahl-Foster methods obtained low results. Klut (27) considered the Schulze-Tiemann method best for the determination of nitric nitrogen in water and sewage, with the Ulsch method as a second choice. Bartow and Rogers (4) compared the aluminum reduction, the phenoldisulphonic acid, and the brucine sulfuric acid methods on water of known nitrate content; none of the results were satisfactory, as indicated by many wide differences in the check determinations of the same method. The aluminum reduction method was then carefully investigated and adopted as best for the water containing chlorides. Burgess (27) investigated the aluminum reduction method, using solutions of known nitrate content, and pronounced the method accurate even in the presence of large quantities of soluble salts and organic matter.

From the above-cited literature it may be seen that none of the methods appear to be wholly without objections, but in so far as their use in the presence of large quantities of soluble salts and organic matter is concerned the

aluminum reduction and the Ulsch methods are best. Therefore, in this work we have investigated these two methods, and on account of its extreme accuracy in the absence of chlorides, sulfates, and soluble organic matter, and because of the fact that the conditions of its use have been so carefully investigated (13), we have used, wherever practicable, the phenoldisulphonic acid method for comparison. The methods as used were therefore the aluminum reduction as modified and described by Burgess (10), the phenoldisulphonic acid method as described by Stewart and Greaves (46), and the Ulsch method as described by Ulsch (51).

In place of the specially constructed pear-shaped glass tube used by Ulsch we have used in the neck of the reduction flask a 2-hole stopper through which passes a bent tube which dips into a vessel containing water. Through the other opening in the stopper is passed a separatory funnel. This prevents the loss of any of the nitrate or ammonium sulfate due to the mechanical action of the rapidly evolved hydrogen. It also makes possible the addition of the sulfuric acid in small quantities, thus insuring complete reduction with smaller quantities of both iron and sulfuric acid than when the acid is all added at once. A series in operation is shown in Plate I.

PREPARATION OF A CLEAR FILTRATE

The investigator of "alkali soils" has difficulty in obtaining a clear filtrate because in the watery extract of such soils we often find the degree of dispersion of the colloidal disperse phase so great and in such a stable form that the solution may be kept for years (7) without flocculation. Furthermore, the particles of such a disperse phase are so small that it is impossible to separate them by means of filtration. It is, however, imperative, especially when using the phenoldisulphonic acid method, that the soil extract be free from colored colloidal material. Still further, no work has been done, so far as we know, to ascertain the influence of these colloids on the various reduction methods. It is quite possible that if the colloidal material be proteinaceous in nature, subsequent treatment may yield from it ammonia which would thus vitiate the results. It is, therefore, essential that we have reliable methods of preparing a clear aqueous solution of the soil nitrates. The ideal method would be one which yielded a clear solution without filtration. This would necessitate the addition of a flocculant to the soil infusion and such a substance should be rapid in action, remove only a minimum quantity of the nitrate from solution and must not interfere with the sensitiveness of the method.

In our search for such an agent we have been guided by two main lines of evidence; first, those substances which have proved most efficient in flocculating other colloids from their dispersing media probably would be most efficient for soil; second, various flocculants have been used and recommended by others workers on similar soils.

The work of Linder and Picton (33) is especially valuable in this connection, as they tested the precipitating powers of various salts on arsenious sulfide solution. Aluminum sulfate was the most efficient compound used, followed by aluminum chloride. Ferric sulfate was only about one-third and calcium salts were only one-fourth as effective, while it required 3200 times as much sodium chloride as aluminum sulfate to bring about the same effect. Furthermore, McGeorge (34) has found aluminum sulfate the most efficient flocculant for clay. But while Wolkoff (58) found the aluminum compounds among the best, there was a great variation, depending upon the soil. This is a fact which may be expected when we recall Hardy's rule that the precipitating power of an electrolyte for a colloid depends on the valence of the ion which carries the electric charge opposite to that of the dispersoid. The colloids found in the soil may be either the electro-negative colloid, clay, or the positive or negative organic colloid.

Buhlert and Fickendey (9) recommended the addition of 2 per cent of sodium chloride, while King and Whitson (26) used potassium alum. But the quantity of alum recommended by them gave with some soils (44) a cloudy solution which had to be filtered before it could be read in the colorimeter. Furthermore, in the presence of sulfates there is a loss of nitrates when the phenoldisulphonic acid method is used. Chamot (12) and co-workers found aluminum cream the best precipitating agent of several tested for organic matter, and that it occasioned no loss of nitrates. But Lipman and Sharp (30) found potassium alum, aluminum cream, and bone black all decidedly unreliable, as they all induced losses of nitrates in the phenoldisulphonic acid method. They found lime to be much more reliable, the losses of nitrates incurred through its use being small. But it must not be used in this method in connection with chloroform, as the double decomposition which takes place under this condition forms chlorides, thus interfering with the accurate determination of nitrates (47). We have used very extensively in our work (46) on soil nitrates the Chamberland-Pasteur filter, which gives a clear solution and occasions only slight losses of nitrates (8).

From the above review it would appear that the most likely flocculants are sodium, potassium, and iron alum, ferric sulfate, lime, finely precipitated calcium carbonate, and talc. Hence, these, together with the Chamberland-Pasteur filter and the centrifuge, were tested on two soils—one a brickyard clay, the other very high in organic matter.

Two grams of each reagent were used in 500 cc. of water containing 100 grams of the soil. The mixtures were shaken for ten minutes in a mechanical shaker and then stood upright. After standing for ten minutes most of the solutions were quite clear and were pipetted off. They were compared, the one with the other, in a colorimeter with the result that a column 100 cm. deep of the lime-treated solution had a turbidity equivalent to a column of the potassium-alum-treated soil 40 cm. deep; the sodium-alum-treated 27 cm.; the ferric-sulfate-treated 26 cm.; and the ferric-alum-treated solution 24 cm. deep.

Both soil-water mixtures, without a flocculant, were clear after centrifuging fifteen minutes at a speed of 1200 revolutions per minute. Soil-water mixtures to which calcium carbonate or talc had been added were not clear after standing several hours.

The quantity of nitric nitrogen in the clear solution was determined by the aluminum reduction, the Ulsch reduction, and the phenoldisulphonic acid methods. The average results obtained for the clay soil are given in table 1, while those for the soil high in organic matter are given in table 2. Determinations were made of the nitric nitrogen in 20 grams of soil, also determinations of the quantity in 20 grams of soil to which had been added 10 mgm. of nitric nitrogen. In place of reporting the results for each separate determination we have reported the average, together with the standard deviation, according to the following formula:

$$E = \pm \sqrt{\frac{S}{n(n-1)}}$$

in which S = the sum of the squares of the difference between each result and the average of all the determinations; and n = the number of determinations in the average.

It was thought unnecessary to use the phenoldisulphonic acid method in the presence of any of the sulfates, as previous workers have shown these substances to interfere with the accuracy of the method.

The results obtained on the soil alone by the Ulsch method are invariably higher than those obtained by either of the other methods. They are also more uniform in the method of Ulsch than in the other methods. Furthermore, as may be seen from an examination of the reported standard deviation, the agreement between duplicate determinations is better than with the other methods. The greatest deviation in the Ulsch method is only ± 0.14 . In the aluminum reduction method it is ± 0.39 , while in the phenoldisulphonic acid method it is ± 1.235 , but in this case the error is great when calcium carbonate was used as the flocculant and the solutions were very turbid when analyzed. But even with allowance for this, the variation between duplicates is much greater than with either of the other methods, for we find that the error is great even where the solution was filtered or centrifuged. These solutions were clear, but the colored soluble colloids which are often in soil solutions, and which it is impossible to remove without oxidation interfere in the phenoldisulphonic acid method.

The average quantity of nitric nitrogen recovered by the Ulsch method where 10 mgm. of nitric nitrogen were added to the soil with the various flocculants was 99 per cent. The minimum quantity (93 per cent) was obtained where the centrifuge was used, and the maximum (101 per cent) where the iron alum was used. The average for the aluminum reduction method was 92 per cent, the minimum (88 per cent) where the talc was used, and the maximum (97 per cent) where the iron alum was the flocculant. The varia-

TABLE I
Milligrams of nitric nitrogen obtained from a heavy clay to which had been added various flocculants

FLOCCULATING AGENT	FILTER	CENTRIFUGE	CALCIUM OXIDE	CALCIUM CARBONATE	TALC	POTASSIUM ALUM	SODIUM ALUM	IRON ALUM	FERRIC SULFATE
<i>Nitric nitrogen found by the aluminum reduction method</i>									
Number of determinations in average.....	4	2	4	4	2	3	3	3	3
Nitric nitrogen from soil.....	0.38	0.64	0.32	0.32	0.32	0.46	0.28	0.25	0.28
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	10.05	9.80	9.49	10.05	9.10	9.85	8.82	9.97	9.10
Nitric nitrogen recovered.....	9.67	9.16	9.16	9.73	8.78	9.39	8.54	9.72	8.82
Standard deviation.....	±0.087	±0.28	±0.104	±0.06	±0.14	±0.04	±0.024	±0.39	±0.031
<i>Nitric nitrogen found by the Ullsch reduction method</i>									
Number of determinations in average.....	3	3	2	4	4	4	4	3	3
Nitric nitrogen from soil.....	0.70	0.78	0.88	0.77	0.83	0.70	0.46	0.35	0.35
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	10.37	10.10	10.51	10.81	10.70	10.37	10.33	10.45	10.08
Nitric nitrogen recovered.....	9.67	9.32	9.63	10.04	9.87	9.63	9.87	10.09	9.73
Standard deviation.....	±0.00	±0.14	±0.00	±0.067	±0.084	±0.029	±0.067	±0.042	±0.084
<i>Nitric nitrogen found by the phenoldisulfophenic acid method</i>									
Number of determinations in average.....	4	3	4	4	4	4	4	3	3
Nitric nitrogen from soil.....	0.16	0.10	0.04	0.06	0.06	0.03			
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	10.87	9.20	13.10	18.28	9.05				
Nitric nitrogen recovered.....	10.71	9.10	13.06	18.22	9.02				
Standard deviation.....	±0.52	±0.30	±0.371	±1.235	±0.21				

TABLE 2
Milligrams of nitric nitrogen recovered from a greenhouse loam soil to which had been added various flocculants

FLOCCULATING AGENT	NITRAS	CENTRIFUGES	CALCIUM OXIDE	CALCIUM CARBONATE	TALC	POTASSIUM ALUM	SODIUM ALUM	IRON ALUM	FERRIC SULFATE
<i>Nitric nitrogen found by the aluminum reduction method</i>									
Number of determinations in average.....	3	4	4	3	2	2	4	3	2
Nitric nitrogen from soil.....	1.92	1.68	2.17	1.86	1.95	1.6	2.21	1.86	2.07
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	11.19	11.76	11.83	11.39	11.00	11.91	11.49	11.50	11.00
Nitric nitrogen recovered.....	9.27	10.08	9.65	9.52	9.05	10.30	9.28	9.62	8.92
Standard deviation.....	±0.20	±0.14	±0.093	±0.16	±0.07	±0.05	±0.03	±0.14	±0.07
<i>Nitric nitrogen found by the Ulisch reduction method</i>									
Number of determinations in average.....	4	3	3	4	3	2	3	3	3
Nitric nitrogen from soil.....	2.26	2.38	2.52	2.70	2.98	2.28	2.45	2.51	2.24
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	12.26	12.44	12.19	12.57	12.98	12.12	12.42	11.98	11.8
Nitric nitrogen recovered.....	10.00	10.06	9.67	9.87	10.00	9.82	9.97	9.37	9.56
Standard deviation.....	±0.09	±0.04	±0.09	±0.07	±0.12	±0.07	±0.02	±0.04	±0.05
<i>Nitric nitrogen found by the phenoldisulphonic acid method</i>									
Number of determinations in average.....	4	4	4	4	4	4	4	4	4
Nitric nitrogen from soil.....	1.77	1.50	2.81	1.79	1.65				
Nitric nitrogen from soil + 10 mgm. nitric nitrogen.....	14.00	10.47	14.65	11.27	10.60				
Nitric nitrogen recovered.....	12.23	8.97	11.84	9.48	8.92				
Standard deviation.....	±0.01	±0.49	±0.33	±0.19	±0.28				

tion in results where the phenoldisulphonic acid method was used is very great, amounting in the extreme to over 100 per cent. It is interesting to note that where the calcium carbonate was used as the flocculant, usually greater quantities of nitrates were recovered than when the other clarifying agents were used. This may be due to the absorption of some of the nitrate by the flocculated colloid or to the precipitant carrying down mechanically some of the nitrates with the colloids, or in the turbid solutions the fine material left in suspension may contain nitrogen compounds which on subsequent treatment yield ammonia. This latter explanation does not seem plausible on examination of the results given in table 2, where we have a loam soil rich in organic matter. In this series the increase where talc or calcium carbonate is used as the flocculant is no greater than it was with the clay soil, yet the suspended nitrogen-carrying colloid and soluble organic matter would be several times greater in the loam than in the clay soil.

The results, as a whole, for the loam soil are similar to those obtained with the clay. The Ulsch reduction method invariably gives the highest results with the least standard deviation. Moreover, the average nitric nitrogen recovered with the Ulsch method was over 99 per cent of the 10 mgm. of nitric nitrogen which had been added to the soil, while the aluminum reduction method recovers only 95 per cent, and the phenoldisulphonic acid method yields 103 per cent; hence, with both soils the best average and individual results have been obtained with the Ulsch reduction method. Further, there would appear to be a number of objections which may be brought against the aluminum reduction method, as proposed by Burgess. The method, as outlined by him, permits of an appreciable loss of ammonia as one can invariably detect the odor of ammonia on removing the stopper from the reduction tube, and, this being the case, there would be a small loss, and the results which have been reported in the preceding tables indicate that this is a measurable quantity. It is necessary that reduction take place in a thermostat where the temperature can be carefully controlled, in order to obtain even fair results, and it requires several hours for complete reduction. Moreover, even under these conditions we often find tubes in which little or no reduction has taken place, owing to coatings which may be on the fresh aluminum (35), or which may deposit from the soil solution. Hence, the Ulsch reduction method would appear to be better suited to the determination of nitric nitrogen in soil than the aluminum reduction method.

There is nothing in these results which would indicate that a greater error is introduced by the use of the Chamberland-Pasteur filter than by the use of a flocculant, and it is certain that where the phenoldisulphonic acid method is to be used the solution should be prepared by centrifuging or filtering through the Chamberland-Pasteur filter. With the Ulsch method results so far reported indicate that quick lime or alum may be permissible, as they give clear solutions with a considerable saving of time.

But Patten and Snyder (37) found that where lime had been used as the

flocculant, both the phenoldisulphonic acid and the aluminum reduction methods gave low results. Moreover, they claim that calcium carbonate is just as efficient in clarifying soil solutions as is lime; but this is not in keeping with our experience, for many of the soil solutions with which we have to deal and to which either calcium carbonate or talc have been added are not clear even after standing several hours. For these reasons we have used the Ulsch method on solutions which have been clarified by both lime and alum to determine the minimum quantity of these flocculants necessary to produce a clear solution and to determine the influence of varying quantities upon the accuracy of the method. The average results from such a test are given in table 3. They are the average of four or more determinations. The determinations were made at different times and on two different soils, hence are not directly comparable the one with the other, but each is comparable where it received different quantities of the respective flocculant. Both were rich greenhouse soils.

TABLE 3

Nitric nitrogen found in 100 grams of soil to which various quantities of lime and alum had been added

QUANTITY OF FLOCCULANT USED grams	MILLIGRAMS OF NITROGEN FOUND IN 100 GRAMS OF SOIL IN THE PRESENCE OF			
	Lime	Standard deviation	Alum	Standard deviation
0.5	19.00	±0.213	9.14	±0.014
1.0	18.30	±0.329	9.17	±0.014
2.0	18.20	±0.403	9.31	±0.042
5.0	18.00	±0.084		

It may be seen from the results reported in tables 1 and 2 that solutions which have been treated with lime gave low results as compared with others which were clarified by the centrifuge or the Chamberland-Pasteur filter, and from the results in table 3 we find that the quantity of nitric nitrogen recovered from a soil varies with the quantity of lime added. Furthermore, as may be seen from the results reported under "standard deviation," the agreement existing between determinations made on samples containing the same quantities of nitric nitrogen and lime do not agree. But, even in the presence of lime, in none of our work with the Ulsch method have we obtained such variation as is reported by Patten and Snyder when the aluminum reduction or phenoldisulphonic acid methods are used. But it is evident that lime does interfere, even in the Ulsch method.

Where alum was used as the flocculant, clear solutions were obtained with 0.5 gram and it is not likely that more than 2 grams would ever be required to clarify the solutions from any soil. The results obtained with this flocculant are slightly high and can readily be obviated by using in the check on the chemicals the same quantity of alum as has been applied to the soil. It is

important to note that the variation between different determinations is very small where alum was used as the flocculant.

The low results obtained in the presence of lime are not due to the removal by the lime of colloidal organic material, which, if left in solution, may in subsequent treatment break down and yield ammonia, for similar low results are obtained when lime is added to a pure solution of sodium nitrate, as may be seen from the results given in table 4.

TABLE 4
Nitric nitrogen recovered from solutions of sodium nitrate containing various flocculants

CONTENTS OF SOLUTION USED	NITRIC NITROGEN FOUND	STANDARD DEVIATION
	mgm.	
40 mgm. nitric nitrogen.....	39.88	± 0.150
40 mgm. nitric nitrogen + 5 grams lime.....	38.84	± 0.599
40 mgm. nitric nitrogen + 2 grams alum.....	39.93	± 0.128
40 mgm. nitric nitrogen + 10 grams CaCO_3	39.09	± 0.420
40 mgm. nitric nitrogen + 10 grams talc.....	39.50	± 0.193

There is nothing in these results which could be taken to indicate that alum interferes in the Ulsch method any more than does the calcium carbonate and talc. But there is a small loss where the lime is used. The variation between determinations is no greater where the alum was used than in its absence, while where the lime has been used the variation between determinations is comparatively large. Hence, it would appear that potassium alum gives a clear solution with a minimum loss of nitric nitrogen and does not appreciably interfere with the accuracy of the method.

TIME OF EXTRACTION

The time necessary for water added to a soil to dissolve the nitric nitrogen within that soil may vary greatly with the condition of the soil when the water is added, as well as the treatment given the soil-water mixture. But if the water be not left in contact with the soil for a sufficient length of time, some particles may not be wet; hence, some of the nitric nitrogen will be left unextracted—if left too long, denitrification may take place. Yet we find a large variation in the time recommended by different workers. King and Whitson (26) extracted the soil in a mortar with continual grinding for two minutes, and this is the time which has been used very extensively by us (46). Buhlert and Fickenday (9) agitated the soil with water for one-fourth of a minute at intervals of five minutes for thirty minutes and then filtered. A similar procedure was used by Gutzeit (20), while others (55) extracted for two hours. Sazanov (40) found that longer contact of the water with the soils than enough to allow the soils to settle leads to a loss of nitrates.

In order to obtain more exact information on this subject we have extracted 100-gram portions of soil containing 2 grams of alum with 500 cc. of water for

varying lengths of time. A rich greenhouse soil was used. It was mixed with the water in 1000-cc. bottles and shaken in a mechanical shaker for time indicated, then allowed to stand long enough to settle—which was not over ten minutes—and the nitric nitrogen determined. The average results from four or more determinations are given in table 5.

TABLE 5
Nitric nitrogen extracted from soil in given time

TIME SHAKEN	MILLIGRAMS OF NITRIC NITROGEN FOUND IN 100 GRAMS OF SOIL	STANDARD DEVIATION
minutes		
5	21.70	±0.02
10	21.70	±0.06
15	21.30	±0.03
30	21.90	±0.03
35	21.90	±0.03
40	21.20	±0.04

These results indicate that nothing is to be gained by shaking the soil with the water longer than five minutes. But it must be borne in mind that these tests were carried out on finely powdered soil and not upon lumps, and, furthermore, it was thoroughly agitated during the time it was in the shaker.

RATIO OF SOIL TO WATER

We find different workers vary the ratio of water to soil. Fricke (17) and Schmidt (42) with fertilizers used one part of sample to fifty parts of water. Litzendorff (31) used one part of soil to two parts of water. Buhlert (9) and Gutzeit (20) used one part of soil to either one or one and one-half parts of water. King and Whitson (26) used one part of soil to five parts of water, and this is the ratio which is used by many workers at the present time. We have made the determinations of nitric nitrogen in soil, and soil to which dried blood was added, using various ratios of water to soil, while all of the other conditions were held the same on all samples. The average results for four or more determinations are given in table 6.

TABLE 6
Nitric nitrogen extracted from soil with various quantities of water

RATIO OF SOIL TO WATER	MILLIGRAMS OF NITRIC NITROGEN IN 100 GRAMS SOIL	STANDARD DEVIATION	MILLIGRAMS OF NITRIC NITROGEN IN 100 GRAMS SOIL CONTAINING 2 PER CENT DRIED BLOOD	STANDARD DEVIATION
1-5	8.93	±0.034	9.80	±0.056
1-10	8.82	±0.034	9.10	±0.052
1-15	8.70	±0.033	9.80	±0.008
1-20	8.75	±0.028	9.45	±0.028
1-25	8.95	±0.029	9.38	±0.052

The difference found is only what could be attributed to experimental error. Hence, we may conclude that the ratio of soil to water is immaterial, provided we do not add sufficient nitric nitrogen to give a saturated solution. But it is important that the water in the sample be taken into consideration. The facts that the same quantity of nitric nitrogen is extracted with the varying quantities of water and that practically 100 per cent of the nitric nitrogen added to the soil is recovered in the various experiments reported in this work, make it very doubtful if Allen and Bonazzi's (2) contentions are justified, that the determination of the nitric nitrogen in the water extract does not give a determination of the nitric nitrogen in the soil.

USE OF AN ANTISEPTIC

The majority of workers assume that if the soil is to stand in contact with the water any length of time, an antiseptic should be added. Yet, we have no definite information on the quantity which should be added in order

TABLE 7
Nitric nitrogen recovered from soil after varying lengths of time

TIME	MILLIGRAMS OF NITROGEN RECOVERED IN					
	100 grams soil	Standard deviation	100 grams soil + 20 mgm. nitric nitrogen	Standard deviation	100 grams soil + 40 mgm. nitric nitrogen	Standard deviation
Nitric nitrogen at first.....	36.85	± 0.03	53.35	± 0.35	72.85	± 0.16
Nitric nitrogen after 24 hours.....	38.53	± 0.03	57.44	± 0.28	74.23	± 0.07
Nitric nitrogen after 48 hours.....	34.00	± 0.02	55.28	± 0.30	70.00	± 0.04
Nitric nitrogen after 120 hours.....	26.00	± 0.10	50.40	± 0.06	67.20	± 0.13
Nitric nitrogen after 144 hours.....	19.90	± 0.02	46.24	± 0.60	53.95	± 0.22

to be effective, and, in many cases, the quantity added may actually accelerate instead of retard or stop the biological transformations going on in the soil. Moreover, it is not immaterial as to what antiseptic be used, for chloroform (47) used in the presence of lime with the phenoldisulphonic acid method gives inconsistent results.

In order to determine the error which may enter, due to the standing of the soil solution, we treated 100-gram portions of a rich garden soil with water and varying quantities of nitric nitrogen and determined the quantity of nitric nitrogen present after varying lengths of time. The average results for a number of determinations are given in table 7.

It is interesting to note that no nitrogen is lost from the solution containing only alum during the first twenty-four hours, and the amount lost during the first forty-eight hours where thymol was used amounts to less than 3 mgm. It must be remembered that this is a soil heavily laden with horse

manure. The solutions to which thymol had been added lost no nitrogen during the first forty-eight hours, but by the end of one hundred and forty-four hours practically all of the nitric nitrogen had disappeared. This may have been due to denitrification, or to the rapid multiplication of other micro-organisms within the solutions which were favored by the thymol and had transformed the nitric nitrogen into protein within their bodies.

Where chloroform was used the loss was very slight, even after one hundred and forty-four hours, but it is interesting to note that where the thymol or chloroform was used the results are higher than in their absence. This is not due to the prevention of bacterial growth, for the results are higher at first in the presence of the antiseptic than in its absence. And we have found this to be the case in many other tests which are not reported here. Furthermore, in the absence of thymol and chloroform where the 20 or 40 mgm. of nitric nitrogen were added, practically 100 per cent was recovered in the three determinations made during the first forty-eight hours, but where the antiseptic was added, especially the chloroform, the quantity recovered is often considerably greater or less than that added.

However, the quantity of nitric nitrogen lost from a mixture of soil and fresh horse manure may be considerable, even where the 2 grams of alum have been used as a flocculant, as is seen by the following results.

	LIME	ALUM
Milligrams of nitric nitrogen found at first.....	48.34	48.94
Milligrams of nitric nitrogen found after 24 hours.....	45.53	41.58
Milligrams of nitric nitrogen found after 72 hours.....	44.50	22.42
Milligrams of nitric nitrogen found after 96 hours.....	45.80	20.30

Here we find no loss of nitric nitrogen during the first twenty-four hours either in the soil solution or in that to which sodium nitrate had been added, and even after forty-eight hours the loss was extremely small. Repeated tests on ordinary fertile soil kept at 24°C. in solutions showed no loss after twenty-four hours and the loss was always very small after forty-eight hours.

That the added antiseptic, if used in too small a quantity, may actually increase bacterial activity and thus increase the loss of nitric nitrogen, may be seen from the results reported in table 8. In this series a soil rich in manure was used; to one set was added only the flocculant—2 grams of alum—to the second 2 grams of alum plus 2 cc. of a saturated solution of thymol, and to the third 2 grams of alum plus 0.5 cc. of chloroform. All were kept at a temperature of 24°C. The results as reported are the average of a number of determinations.

While in this case we are dealing with solutions with optimum conditions for denitrification, the loss in the presence of lime is small, but in the case of the alum it is appreciable. This, however, is a soil extremely rich in nitric nitrogen and the results probably represent a maximum denitrification, and

TABLE 8
Nitric nitrogen in soil after varying lengths of time

TREATMENT	AT FIRST	STANDARD DEVIATION	AFTER 24 HOURS	STANDARD DEVIATION	AFTER 48 HOURS	STANDARD DEVIATION	AFTER 144 HOURS	STANDARD DEVIATION	AFTER 168 HOURS	STANDARD DEVIATION
Soil + 2 grams alum.....	48.00	± 0.03	47.63	± 0.025	45.20	± 0.04	35.00	± 0.09	33.60	± 0.00
Soil + 2 grams alum + 20 mgm. nitric nitrogen.....	67.96	± 0.05	67.26	± 0.025	65.44	± 0.03	58.14	± 0.09	55.34	± 0.05
Soil + 2 grams alum + 40 mgm. nitric nitrogen.....	89.68	± 0.06	87.28	± 0.033	84.34	± 0.06	77.01	± 0.05	72.85	± 0.57
Soil + 2 grams alum + thymol.....	49.04	± 0.05	49.00	± 0.07	48.30	± 0.10	4.00	± 0.22	5.00	± 0.57
Soil + 2 grams alum + thymol + 20 mgm. nitric nitrogen.....	69.36	± 0.05	67.68	± 0.03	67.96	± 0.10	4.40	± 0.19	4.00	± 0.05
Soil + 2 grams alum + thymol + 40 mgm. nitric nitrogen.....	89.26	± 0.05	88.26	± 0.068	8.26	± 0.04	1.40	± 0.35	4.00	± 0.00
Soil + chloroform.....	51.80	± 0.10	51.80	± 0.03	51.30	± 0.04	43.40	± 0.04	37.80	± 0.00
Soil + chloroform + 20 mgm. nitric nitrogen.....	71.94	± 0.08	71.87	± 0.09	71.46	± 0.05	60.66	± 0.11	57.55	± 0.07
Soil + chloroform + 40 mgm. nitric nitrogen.....	88.46	± 0.11	91.06	± 0.04	88.26	± 0.14	80.84	± 0.30	72.86	± 0.17

such activity would never be reached with average soil. But even this loss can be prevented by the use of chloroform, as may be seen from table 9 which shows the quantities of nitric nitrogen recovered from a soil manure mixture in water at 24°C. after varying lengths of time.

By the end of three hours the soil to which no antiseptic had been added had lost one-half of its nitric nitrogen, while during twenty-four hours it had been reduced to 0.83 mgm. The soil to which the 2 grams of alum only had been added slowly lost its nitric nitrogen so that after seventy-two hours its amount had been reduced to one-half that present at the beginning. Where the chloroform was added the quantity remains fairly uniform through-

TABLE 9
Nitric nitrogen from soil and manure after varying lengths of time

TREATMENT	MILLIGRAMS OF NITRIC NITROGEN FOUND			
	In beginning	After 24 hours	After 48 hours	After 72 hours
Soil, no antiseptic.....	3.064	0.826	0.49	0.56
Soil + 2 grams alum.....	3.117	2.732	2.59	1.96
Soil + 2 grams alum + 0.5 cc. CHCl_3	3.067	2.980	3.06	3.22
Soil + 2 grams alum + 1.0 cc. CHCl_3	2.956	2.840	2.90	2.91
Soil + 2 grams alum + 2.0 cc. CHCl_3	2.800	3.010	2.90	2.89
Soil + 2 grams alum + 3.0 cc. CHCl_3	2.790	2.910	2.93	3.16

out the seventy-two hours and it would appear that 0.5 cc. of chloroform is ample to prevent loss of nitric nitrogen. But it has been our experience that where the analyses are completed immediately, more concordant results are obtained without than with chloroform. The average results on both, rich soil and a soil manure mixture, are shown graphically in figure 1. In this the quantity of nitric nitrogen in the soil at the beginning of the experiment has been considered as 100 per cent.

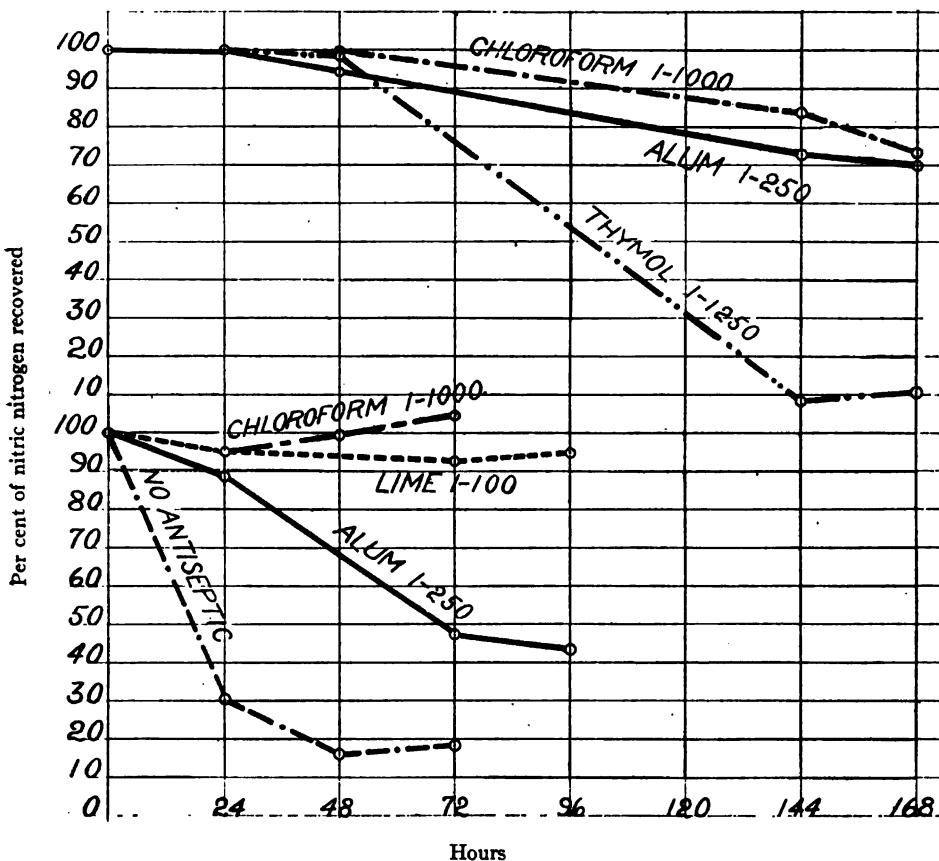


Fig. 1 Curves showing per cent of nitric nitrogen recovered in sample of soil treated by different antiseptics after varying lengths of time.

The soil with 2 grams of alum during the first twenty-four hours lost 0.8 per cent of its nitric nitrogen, while those to which chloroform or thymol had been added lost none. By the end of one hundred and sixty-eight hours the soil plus 2 grams of alum had lost 30 per cent, the soil with the chloroform had lost 27 per cent, while that to which the thymol had been added had lost 89.8 per cent of its nitric nitrogen. The loss from the horse manure soil

mixture—with no antiseptic—during the first twenty-four hours was 73 per cent, but it was very low in the presence of the antiseptics.

From all the results reported it would appear that where the alum is used as a flocculant no other antiseptic is necessary unless the solutions are to remain several hours. The losses of nitrogen reported in most of this work are maxima and probably would never be duplicated in regular work, as the soil contained such large quantities of nitrates and organic matter of fresh horse manure. Moreover, the solutions were kept in an incubator at 24°C. during various intervals. Hence, it would appear that, under ordinary conditions, where the time in which the soil is in the water is short, and the temperature comparatively low, no loss would result with ordinary soil where the 2 grams of alum had been added.

QUANTITY OF ACID AND IRON

Ulsch (50) in his original method recommends that the nitrate-carrying substance be dissolved in 25 cc. of water, and this reduced with 5 grams of

TABLE 10

Nitric nitrogen from 20 grams of soil plus 40 mgm. of nitric nitrogen with different quantities of sulfuric acid and iron

IRON USED grams	H ₂ SO ₄ USED cc.	NITRIC NITROGEN USED mgm.	STANDARD DEVIATION	PERCENTAGE RECOVERED
5	10	35.76	±0.51	82.62
5	20	40.21	±0.30	92.90
5	25	42.45	±0.58	98.08
5	30	42.90	±0.10	99.10
6	20	42.73	±0.31	98.72
6	25	42.24	±0.50	97.60
6	30	42.59	±0.06	98.40

iron and 10 cc. of dilute sulfuric acid (1 to 2). Later (51) he found 10 grams of iron and 10 cc. of dilute sulfuric acid (sp. gr. 1.35), sufficient to reduce 0.5 gram of potassium nitrate. Schenke (41) recommends 4 grams of iron and 12 cc. of sulfuric acid (sp. gr. 1.35). Work at the New Jersey Station (3) indicates that 2 grams of iron and 5 cc. of sulfuric acid was sufficient for 1-gram samples of any fertilizer, while the Street (48) modification requires 2 or 3 grams of reduced iron and 10 cc. of sulfuric acid (one part of acid to one of water). Our experience has been that the best results are obtained when the ratio between the iron and acid is large, as is brought out by the results in table 10. In this work the quantity of nitric nitrogen determined was that extracted from 20 grams of a rich soil to which had been added 40 mgm. of nitric nitrogen. The results are the average of four or more determinations. Repeated determinations had yielded from 20 grams of the soil 3.28 mgm. of nitric nitrogen; hence, the quantity of nitric nitrogen actually placed in the reduction flask was 43.28 mgm.

Here we find the greatest quantity of the nitric nitrogen was recovered where 5 grams of iron were used with 30 cc. of sulfuric acid (sp. gr. 1.35). The results vary with the method of adding the acid, the reduction being, more complete when the acid is slowly added by means of a dropping funnel as previously outlined. But in no case were we able to obtain concordant results when the quantity of acid used was such as to leave considerable undecomposed iron in the reduction flask.

The quantity of iron necessary will vary with the quantity of nitric nitrogen to be reduced and also with the grade of iron used, as may be seen from the results reported in table 11. Here we have used three different grades of iron and 25 cc. of sulfuric acid with the soil and nitrates as in previous experiments.

The "iron-by-hydrogen 90 per cent" yielded the same quantity of nitric nitrogen when 7 grams were used as when 5 grams were used. But the 80 per cent and the alcoholized iron both yielded more nitric nitrogen with the

TABLE 11
Nitric nitrogen recovered with various grades and quantities of iron

KIND OF IRON	IRON USED grams	NITRIC NITROGEN RECOVERED mgm.	STANDARD DEVIATION	PERCENTAGE OF NITRIC NITROGEN RECOVERED mgm.
Iron by hydrogen, 90 per cent.....	5	42.80	±0.11	98.90
Iron by hydrogen, 90 per cent.....	7	42.69	±0.07	98.52
Iron by hydrogen, 80 per cent.....	5	41.92	±0.22	96.61
Iron by hydrogen, 80 per cent.....	7	42.95	±0.14	99.00
Alcoholized iron.....	5	40.03	±0.17	90.20
Alcoholized iron.....	10	42.27	±0.25	97.05

larger quantities of iron. This is probably due to 5 grams of either 80 per cent or alcoholized reagent not containing enough elementary iron for the complete reduction of 40 mgm. of nitric nitrogen. None of the samples of iron tested contained impurities which would yield ammonia, for repeated tests failed to reveal the presence of ammonia, ammonium compounds, or nitric nitrogen; nor did these samples of iron contain a reducible organic compound as reported by Brandt (6) and later by Hartwell and Wheeler (21).

Moreover, it is certain that 5 grams of 90 per cent "iron-by-hydrogen" is ample for the reduction of 40 mgm. of nitric nitrogen, but where lower grades are used more will be necessary. Even when 10 grams of the alcoholized iron were used we failed to recover all of the nitric nitrogen, and it would not be practicable to use larger quantities, as it introduces many difficulties in technique.

TIME NECESSARY FOR REDUCTION

The time required for complete reduction varies with the temperature. Ulsch (50) found that the percentages of nitric nitrogen recovered when

0.5-gram portions of potassium nitrate were reduced at varying temperatures with appropriate quantities of iron and sulfuric acid were as follows:

	Per cent reduced
20°C. for two hours.....	95
30°C. for two hours.....	97
40°C. for two hours.....	100
90°C. for eight minutes.....	99.5
100°C. for two minutes.....	98.9

This is in keeping with the results obtained by us where 20 mgm. of nitric nitrogen were reduced at varying temperatures.

The results shown in table 12 would indicate that the reduction is not complete even after four hours at the lower temperature, but that when the solution is boiled all of the nitric nitrogen is recovered. It is important to

TABLE 12
Percentages of nitric nitrogen recovered in various reductions

TEMPERATURE	TIME	PERCENTAGE OF NITRIC NITROGEN RE- COVRED	STANDARD DEVIATION
		hours	
25°C.....	2	93.00	±0.58
50°C.....	2	94.50	±0.15
25°C.....	3	91.40	±0.62
50°C.....	3	96.65	±1.00
25°C.....	4	97.70	±0.12
50°C.....	4	98.75	±0.03
Cold $\frac{1}{2}$ hour. Boiled until hydrogen ceased.....		99.75	±0.04
Cold, then boiled ten minutes.....		99.75	±0.04

note that the variation between the determinations is very great when the temperature is low and the time given for reduction short, but where the reduction was conducted at 50°C. for four hours, or by boiling the solution, the agreement was very good. Therefore, it would seem to be advisable to finish the reduction at boiling temperatures and to return the liquid carried over mechanically to the reduction flask before the evolution of hydrogen has ceased, so as to insure reduction of any nitric nitrogen which may have been carried over by the first rapid evolution of hydrogen.

INFLUENCE OF VARIOUS SALTS

Many of the solutions obtained from the arid and semi-arid soils are very rich in soluble salts; hence, it is necessary that the method used for the determination of nitric nitrogen be reliable even when used on solutions containing large quantities of soluble salts. Therefore, we have made determinations using solutions to which had been added various salts which may occur in the soil solutions. These naturally fall into three series treated as follows:

The first series contained 20 mgm. of nitric nitrogen in 500 cc. of water; the second, 20 mgm. of nitric nitrogen and 5 grams of one of the salts given in table 13 in 500 cc. of water; the third series contained 20 mgm. of nitric nitrogen, 5 grams of one of the salts, and 100 grams of soil and the 500 cc. of water. The soil used in this last series, as an average of ten determinations, had yielded 0.728 mgm. of nitric nitrogen from 100 grams of soil.

The average quantity of nitric nitrogen recovered from the nitric nitrogen solution was 98.5 per cent, exactly the average quantity recovered where the

TABLE 13
Milligrams of nitric nitrogen recovered from various solutions

SOLUTION CONTAINING 20 MGM. OF NITRIC NITROGEN	STANDARD DEVIATION	SALT ADDED	20 MGM. NITRIC NITRO- GEN PLUS 1 PER CENT SALT	STANDARD DEVIATION	20 MGM. NITRIC NITRO- GEN PLUS 100 GRAMS SOIL PLUS 1 PER CENT SALT	STANDARD DEVIATION
20.16	± 0.00	NaCl	19.80	± 0.03	20.36	± 0.10
19.62	± 0.00	Na ₂ SO ₄	19.60	± 0.00	20.46	± 0.17
19.82	± 0.03	Na ₂ CO ₃	20.10	± 0.14	19.98	± 0.12
19.47	± 0.04	KCl	19.47	± 0.03	19.75	± 0.00
19.79	± 0.03	K ₂ SO ₄	19.58	± 0.09	20.07	± 0.03
19.61	± 0.13	K ₂ CO ₃	19.79	± 0.03	20.50	± 0.09
19.71	± 0.01	Ca Cl ₂	19.93	± 0.07	20.07	± 0.03
19.71	± 0.05	CaSO ₄	19.79	± 0.07	20.27	± 0.17
19.61	± 0.14	CaCO ₃	19.47	± 0.08	20.18	± 0.08
19.79	± 0.09	MgCl ₂	19.61	± 0.06	20.18	± 0.06
19.65	± 0.07	MgSO ₄	19.79	± 0.09	20.03	± 0.09
19.71	± 0.05	MgCO ₃	19.82	± 0.09	20.13	± 0.05
19.55	± 0.04	MnCl ₂	19.57	± 0.05	20.13	± 0.00
19.68	± 0.04	MnSO ₄	19.75	± 0.00	20.25	± 0.03
19.60	± 0.06	MnCO ₃	19.63	± 0.03	20.09	± 0.03
20.13	± 0.04	FeCl ₃	20.12	± 0.05	20.02	± 0.05
19.88	± 0.12	Fe ₂ (SO ₄) ₃	19.78	± 0.17	19.22	± 0.17
20.10	± 0.09	Na ₂ CO ₃ + Na ₂ O ₄	19.99	± 0.05	20.17	± 0.05

salt had been added to the solution. Where the soil, nitric nitrogen and salt were added, 98.8 per cent of the nitric nitrogen was recovered. Therefore, not one of the seventeen salts added has interfered in this method, for we find the quantity of nitric nitrogen recovered in presence of the salts, both in the pure solutions and with the soil, to be within experimental error the same as that obtained from sodium nitrate solution.

Further, the agreement among determinations is just as good as in the absence of the salt. Moreover, these results give a fair idea of the accuracy of the method.

INFLUENCE OF OTHER SUBSTANCES

Other substances which occur in soil and which may interfere are the soluble organic nitrogenous and non-nitrogenous constituents of soils and manure. The results obtained in the presence of a number of organic constituents are given in table 14.

Both urea and glycocoll yield their nitrogen as ammonia in the Ulsch method, but, as may be seen from the above results, this error may be obviated without a loss of nitric nitrogen by evaporating the alkaline solution to dryness before reduction. In many tests it has been our experience that when dried blood was added to the soil with a definite quantity of nitric nitrogen, slightly less than 100 per cent of the added nitric nitrogen was recovered. There can be but little doubt that this is due to the reacting of the sodium or

TABLE 14
Nitric nitrogen obtained from various organic substances

TREATMENT	NITRIC NITROGEN	STANDARD DEVIATION
		mgm.
0.5 grams peptone	0.04	± 0.17
0.5 grams asparagin (Merks)	0.09	± 0.05
0.5 grams asparagin (Merks) + 10 mgm. nitric nitrogen	10.02	± 0.00
0.5 grams hippuric acid (Merks)	0.07	± 0.07
0.5 grams hippuric acid (Merks) + 10 mgm. nitric nitrogen	9.96	± 0.17
0.5 grams urea	55.65	± 10.30
0.5 grams urea + 10 mgm. nitric nitrogen	90.86	± 3.43
0.5 grams urea + 10 mgm. nitric nitrogen boiled to dryness with 2 cc. NaOH	10.00	± 0.73
10 grams dried blood + 10 mgm. nitric nitrogen	8.89	± 0.14
0.5 grams glycocoll + 10 mgm. nitric nitrogen boiled to dryness with 2 cc. NaOH	9.93	± 0.00

potassium nitrate with the proteins of the added blood. If the resulting compound is insoluble, it will settle from the supernatant liquid and thus be lost; while if a soluble compound is formed, subsequent reduction will fail to liberate the nitric nitrogen. If this is the true explanation of the loss noted we can expect no better results with any of the other methods yet in use.

It is important to note in this connection that the disparate results, reported by some in nitrification experiments where large quantities of dried blood have been used, may, in a measure, be due to this loss of nitric nitrogen noted. However, it is likely that in the regular nitrification experiments, where only 1 or 2 per cent of dried blood has been added to the soil and this incubated for twenty-one days, the error would be very small.

Experiments in which various quantities of artificial humus were mixed with varying quantities of nitric nitrogen showed that this addition did not interfere with the accuracy of the test. Ulsch (52) modified his original method

by using just a little less than enough acid in the receiver to neutralize the ammonia distilled over and then titrating back the excess of ammonia with a standard acid, but we have always used an excess of acid, for Hopkins (24) has pointed out that otherwise there may be a loss of ammonia.

SUMMARY

Clear soil extracts may be obtained by adding 2 grams of lime, ferric sulfate, ferric alum, sodium alum, or potassium alum to the soil-water mixture, by filtering through the Chamberland-Pasteur filter, or centrifuging. The last three procedures give a clear solution with a minimum loss of nitric nitrogen.

Calcium oxide added to a soil-water mixture gives a clear supernatant solution, but the quantity of nitric nitrogen obtained from such a solution is low. This is not entirely due to the mechanical removal of the nitric nitrogen with the precipitated colloid, for when calcium oxide is added to a clear sodium nitrate solution similar results are obtained.

The same quantity of nitric nitrogen was obtained from soil when the ratio of soil to water varied from as low as 1 to 5 to as high as 1 to 25.

No increase in nitric nitrogen is obtained by shaking a soil with water more than five minutes, provided the soil is well pulverized and thoroughly agitated during this time.

Very little nitric nitrogen is lost during twenty-four hours from soil-water mixtures to which 2 grams of alum or 5 grams of lime have been added, and no loss was noted from such solutions even after one hundred and sixty-eight hours when one part per thousand of chloroform had been added to the mixture. But when thymol had been added to such a solution, it accelerated the loss of nitric nitrogen.

None of the following salts interfere in the Ulsch reduction method: chlorides, sulfates, or carbonates of sodium, potassium, calcium, magnesium, manganese and iron. Hence, none of the inorganic salts which may occur in the soil interfere.

When urea or glycocoll are present, high results are obtained unless the alkaline solution is evaporated to dryness before reduction. This may be done without a loss of nitric nitrogen.

When dried blood is mixed with a nitrate less than 100 per cent of the nitric nitrogen is recovered by this method. There can be but little doubt that this is due to the reacting of nitrate with the proteins of the blood. If the resulting compound is insoluble it will settle from the supernatant liquid and thus be lost, while if soluble subsequent reduction will fail to liberate the nitric nitrogen; hence, we would find the same error entering with any of the other methods.

The aluminum reduction method, as modified by Burgess for the determination of nitric nitrogen in alkali soil, permits of an appreciable loss of nitric nitrogen. This is obviated by the use of the iron reduction method which may be outlined as follows.

One hundred grams of the finely ground soil, together with 500 cc. of distilled water, are placed in quart Mason jars and agitated for five minutes—preferably in a shaker. The solution is clarified by one of the following methods:

1. The addition of 2 grams of alum with the soil.
2. Filtering through the Chamberland-Pasteur filter.
3. Centrifugation.

If the alum is used, no other antiseptic is necessary, but unless the analysis is to be completed at once in either of the other cases it is well to add 0.5 cc. of chloroform to each sample. An aliquot part (100 cc.) of the supernatant liquid is pipetted off, and, together with 2 cc. of a saturated solution of sodium hydroxide, evaporated to about one-fourth of its original volume to free from ammonia. If urea is present, it is necessary to evaporate to dryness. To this is added 50 cc. of ammonia-free water, 5 grams of "iron-by-hydrogen" and 30 cc. of sulfuric acid (sp. gr. 1.35). If less than 40 mgm. of nitric nitrogen is to be determined, it is well to take a correspondingly smaller quantity of iron and sulfuric acid. The neck of the reduction flask is fitted with a 2-hole stopper through which passes a 50-cc. separatory funnel and a bent tube which dips into a vessel containing water to prevent mechanical loss. The acid is slowly added and allowed to stand until the rapid evolution of hydrogen is over and then heated to boiling for ten minutes. The contents of the side vessel should be returned to the reduction flask before the reaction is complete, thus insuring the complete reduction of any nitrates which may have been carried over with the first violent evolution of the hydrogen. The contents of the reduction flask are transferred to a Kjeldahl flask, neutralized with sodium hydroxide, and distilled into standard acid. The excess of acid is titrated back with standard alkali, lacmoid being used as an indicator; checks should be made on all the reagents, including the alum used as a flocculant.

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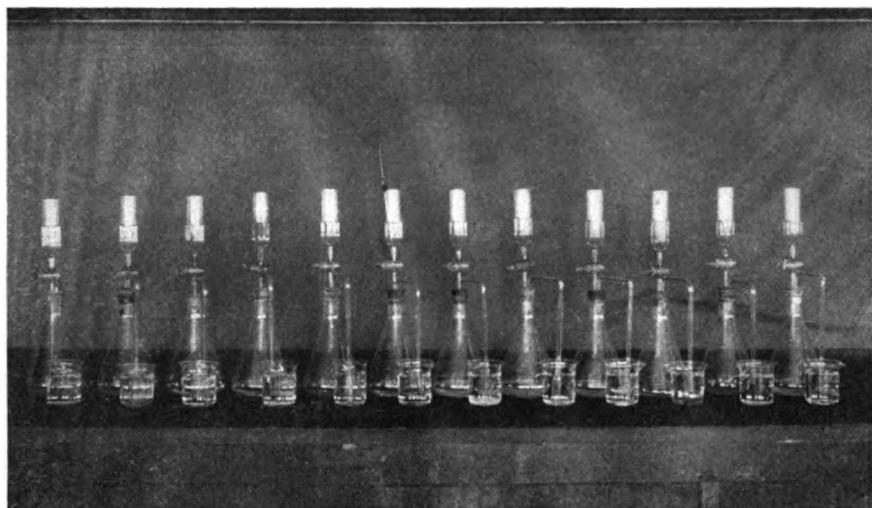
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PLATE 1
APPARATUS USED IN EXPERIMENT SHOWN IN OPERATION

DETERMINATION OF NITRIC NITROGEN IN THE SOIL
J. E. GREAVES AND C. T. HIRST

PLATE 1



THE EFFECTS OF ALKALI SALTS ON NITRIFICATION

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During the last few years it has been recognized that the effects of certain so-called "alkali" salts on soil bacteria, as well as their action on higher plants, should have an important bearing on the development of methods of reclamation of the alkali soils of the arid regions. It was for the purpose of throwing more light on this problem that the following experiments were carried out.

Many experiments have been conducted to determine the concentration at which various alkali salts become toxic to plants, but it remained for C. B. Lipman (8, 9, 10) to point out the importance of bacteriological studies along such lines. The investigations of this author have indicated that bacterial activities in alkali soils are very different from those in normal soils, and that the amount and character of the salts present determine the kind and extent of bacterial action occurring in such soils.

He studied the influence of sodium chloride, sodium sulfate and sodium carbonate on ammonification, nitrification and nitrogen-fixation. He found that all these processes are inhibited by the presence of certain amounts of these salts but the concentration at which each became toxic varied among the different processes. Thus the ammonifying and nitrifying bacteria were differently affected by the salts. The nitrifying bacteria were affected in a manner similar to that shown with the higher plants, but such was not the case with the ammonifying organisms. The nitrogen-fixing organisms seemed to be more resistant to injury from the salts than either of the other groups studied. The results with nitrification which are of special interest in this work showed that sodium carbonate became toxic at a concentration of 0.025 per cent, sodium sulfate at a concentration of 0.35 per cent and sodium chloride at a concentration of 0.1 per cent.

The toxic effects of salts on nitrification have been considered in few other experiments. Deherain (2, 3) noted an injurious effect of sodium chloride on nitrification and J. G. Lipman (11) found that sodium chloride became toxic at 0.1 per cent but did not entirely stop nitrification. This latter experiment was carried out by the solution method.

Headden (5, 6) and Sackett (12) have assumed the occurrence of a very intense nitrification in certain areas in Colorado to account for the excessive accumulations of nitrates occurring there, but Stewart (13) insists that such accumulations are not due to rapid *present* bacterial action, but to a concentration of the nitrates already present in the soils, the irrigation water merely furnishing a medium by which they are moved from place to place.

Furthermore, he asserts that these accumulations of nitrates always occur with large amounts of other water-soluble salts. The study of the effect of salts on nitrification may throw some light on this controversy.

Recent experiments by Kelley (7) on nitrification in semi-arid soils showed that 0.05 per cent of Na_2CO_3 was distinctly toxic to the nitrification of 1 per cent of dried blood, but as high a concentration as 0.4 per cent had no effect on the nitrification of 0.1 per cent of dried blood. When 0.15 per cent of ammonium sulfate was employed in the tests, 0.1 per cent of Na_2CO_3 became toxic to nitrification, but the same amount was markedly stimulating when 0.0625 per cent of ammonium sulfate was used. Similar results were secured with Na_2SO_4 .

No other investigations of a nature to be of value to the general problem have been carried out so far as the authors are aware.

EXPERIMENTAL

The purpose of the experiments reported in the following pages was to determine the concentration at which various alkali salts become toxic to the nitrifying bacteria. Alkali salts, in the concentration which was found in an alkali soil, according to chemical analysis, were added to a similar normal soil. Thus artificial alkali soils containing single salts and various combinations of the same salts were prepared. Comparative tests were run on a typical alkali soil and it was desired by this plan to ascertain the relative and absolute toxicity to the nitrifying bacteria of various salts in actual concentrations found in the field.

Alkali soil was then treated with sodium carbonate and sodium bicarbonate with and without the amount of gypsum necessary to react with the salt added in order to study nitrification under such conditions.

Samples of alkali soil and of a similar normal soil were secured through the courtesy of the Agricultural Experiment Station of the University of Wyoming, and served as a basis for the work. The tests were therefore carried out with typical alkali soil and a typical normal soil from the same region.

Chemical analyses were made of the soils and the water-soluble calcium, sodium, magnesium, potassium, bicarbonate, carbonate, chloride and sulfate determined.

The results of the analyses were as follows:

	NORMAL SOIL per cent	ALKALI SOIL per cent
Calcium (Ca).....	0.0220	0.0550
Magnesium (Mg).....	0.0064	0.0096
Sodium (Na).....	0.0796	0.2570
Potassium (K).....	0.0218	0.0470
Bicarbonate (HCO_3).....	0.0042	0.1190
Chloride (Cl).....	0.0003	0.0084
Sulfate (SO_4).....	0.0980	0.8330
Nitrogen (Total N).....	0.1170	0.0440

The laboratory tests were carried out in tumblers with both the normal and the alkali soils according to the treatments indicated above. One-hundred-gram quantities of the air-dried, sieved soil were weighed out, salt additions made according to the various series, 100 mgm. of ammonium sulfate added, the moisture content of the soils adjusted to the optimum, 5 cc. of a five-minute infusion of a fresh soil added to each soil to introduce a vigorous nitrifying flora, the tumblers were then covered and incubated for four weeks at room temperature, the moisture content being kept up by additions of sterile water to weight every ten days. The nitrates present were then determined by the aluminum reduction method, modified somewhat from the procedure recommended by Burgess (1).

Series I. The effects of NaCl on nitrification in normal soil

The arrangement of this series and the results are shown in table 1 and figure 1. All the treatments were in duplicate and the untreated normal soil and

TABLE 1
Effects of NaCl on nitrification in normal soil

NUMBER	SOIL	NaCl	N	AVERAGE N
			grams	mgm.
1	Normal	None	2.1728	
2	Normal	None	2.2316	2.2022
3	Normal	0.005	3.1164	
4	Normal	0.005	3.2460	3.1812
5	Normal	0.010	2.2316	
6	Normal	0.010	2.5452	2.3884
7	Normal	0.020	2.0328	
8	Normal	0.020	1.7164	1.8746
9	Normal	0.040	0.6608	
10	Normal	0.040	0.9464	0.8536
11	Alkali	None	2.0888	
12	Alkali	None	1.8312	1.9600

untreated alkali soil were used in this and all succeeding series for the sake of comparison.

The addition of 0.005 per cent of NaCl gave a stimulation in nitrate production but the large amounts, at least beyond 0.010 per cent depressed nitrification. This latter amount apparently had no effect.

The alkali soil gave a smaller nitrification than the normal soil and about the same as that receiving 0.020 per cent of NaCl. The toxic point for the NaCl in this test evidently occurred at about 0.02 per cent, which is very much lower than the concentration at which Lipman found toxicity. Differences in the soils tested may account for the variation in the results. It is apparent, however, that small amounts of NaCl stimulate nitrification, while larger amounts bring about a depression.

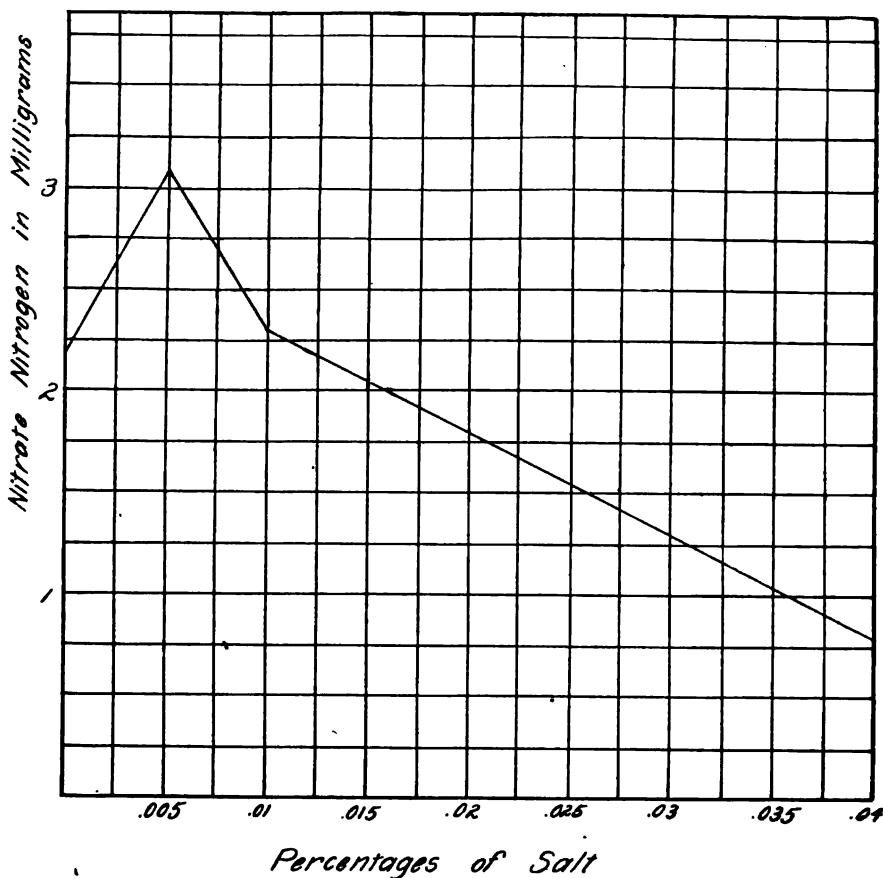


FIG. 1. EFFECTS OF NaCl ON NITRIFICATION IN NORMAL SOIL

Series II. The effects of Na₂SO₄ on nitrification in normal soil

This series was arranged in a manner similar to the preceding except that the treatments with Na₂SO₄ were much larger than those with NaCl. The data obtained are presented in table 2 and figure 2.

The addition of the Na₂SO₄ up to 2.0720 per cent increased considerably the nitrifying power of the soil. The greatest increase was secured with 0.5180 per cent of the salt, but the differences between the results with this application and with the larger amounts were small until 4.1440 per cent was added, when a distinct depression in nitrification occurred. The concentration at which the sodium sulfate becomes toxic was therefore much higher in this soil than in Lipman's experiments, where the toxic point was found to be 0.35 per cent. The variation in the soil used would again probably explain the difference in the results. It is evident again, however, that sodium

TABLE 2
Effects of Na₂SO₄ on nitrification in normal soil

NUMBER	SOIL	Na ₂ SO ₄ grams	N	AVERAGE N
			mgm.	mgm.
1	Normal	None	3.3150	
2	Normal	None	3.6960	3.5030
3	Normal	0.5180	4.6280	
4	Normal	0.5180	Lost	4.6280
5	Normal	1.0360	4.4570	
6	Normal	1.0360	4.3140	4.3850
7	Normal	2.0720	4.1740	
8	Normal	2.0720	3.8860	4.0290
9	Normal	4.1440	1.6010	
10	Normal	4.1440	1.7750	1.6880
11	Alkali	None	3.1720	
12	Alkali	None	2.9450	3.0580

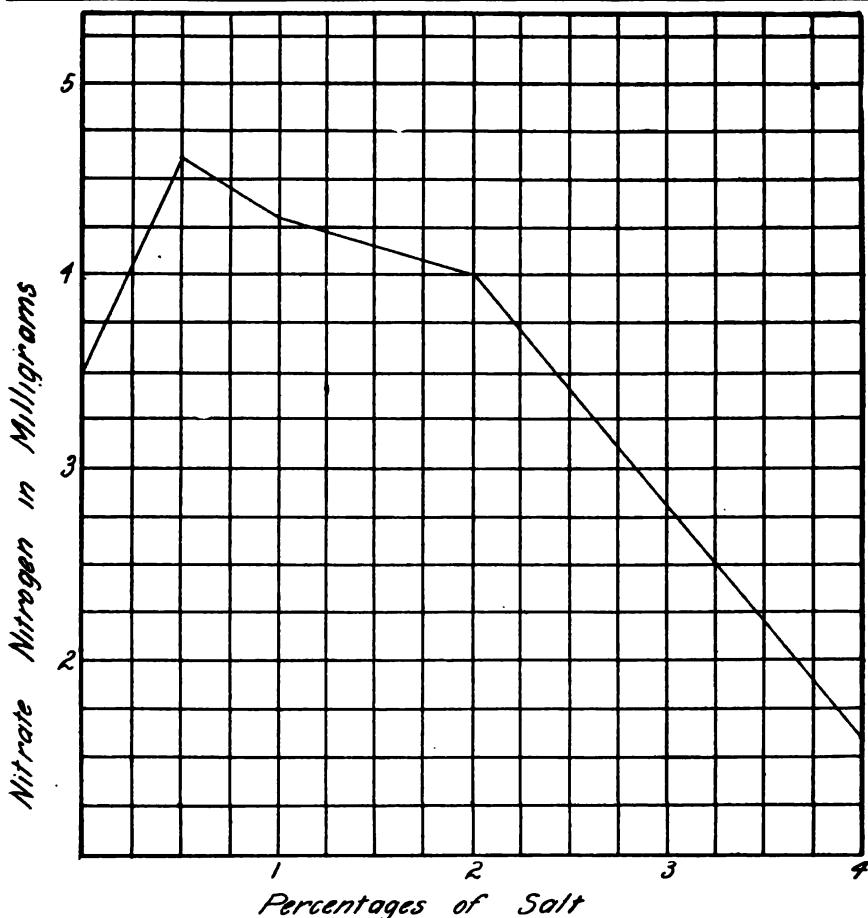


FIG. 2. EFFECTS OF Na₂SO₄ ON NITRIFICATION IN NORMAL SOIL

sulfate stimulates nitrification when added in small amounts, but when the concentration is increased beyond a certain point, which varies in different soils, it becomes distinctly toxic to the nitrifiers.

Series III. The effects of MgSO₄ on nitrification in normal soil

The arrangement of this series and the results secured appear in table 3 and figure 3.

TABLE 3
Effects of MgSO₄ on nitrification in normal soil

NUMBER	SOIL	MgSO ₄	N	AVERAGE N
		grams	mgm.	mgm.
1	Normal	None	2.5375	
2	Normal	None	2.4841	2.5108
3	Normal	0.0235	4.0984	
4	Normal	0.0235	4.2998	4.1991
5	Normal	0.0570	3.4020	
6	Normal	0.0570	4.0292	3.7156
7	Normal	0.1140	3.6008	
8	Normal	0.1140	3.6306	3.6162
9	Normal	0.2280	3.1724	
10	Normal	0.2280	3.0604	3.1164
11	Alkali	None	2.0964	
12	Alkali	None	2.0952	2.0976

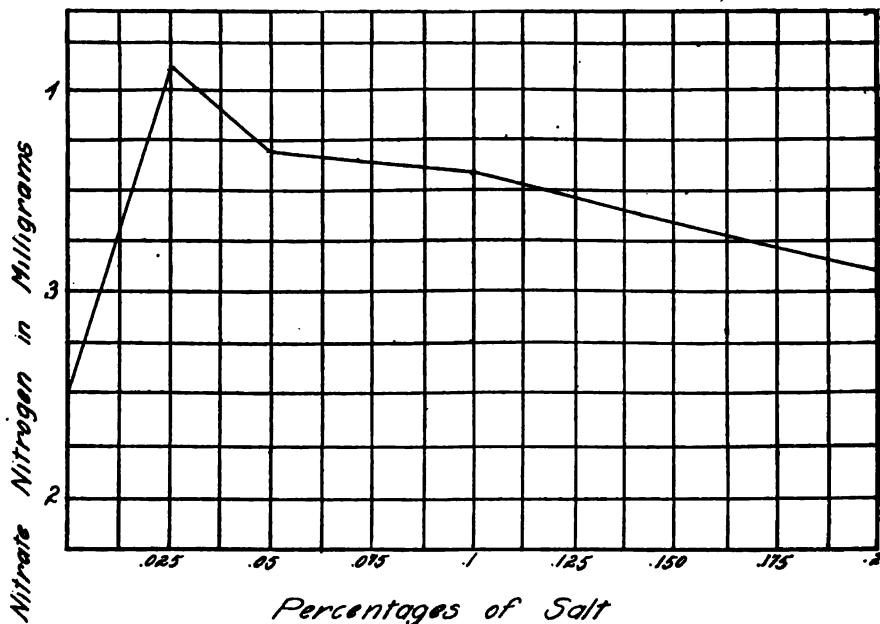


FIG. 3. EFFECTS OF MgSO₄ ON NITRIFICATION IN NORMAL SOIL

The $MgSO_4$ evidently stimulated nitrification in all the amounts used, that is, up to 0.2280 per cent. The greatest stimulation occurred, however, with the smallest application—0.0235 per cent, and the larger amounts brought about smaller effects. If the application had been increased beyond 0.2280 per cent it seems that a depression in nitrification would have occurred, for the figures gradually decreased with increasing applications. Just where a toxic effect from the $MgSO_4$ would occur, cannot, of course, be stated from these results, but it is probable that about 0.3 per cent would mark the point at which depression would occur.

Again it is seen that this salt, like the others tested, stimulates nitrification when present in small amounts, but beyond a certain maximum concentration undoubtedly becomes toxic.

Series IV. The effects of $CaCO_3$ on nitrification in normal soil

The effects of calcium carbonate upon crops and soils have commonly been found to be beneficial, with but few exceptions. It might be expected, therefore, that the nitrifying bacteria would be stimulated in action by additions of this salt.

Examining the results of this series in table 4 and figure 4, it will be seen that a stimulation in nitrification occurred when calcium carbonate was ap-

TABLE 4
Effects of $CaCO_3$ on nitrification in normal soil

NUMBER	SOIL	$CaCO_3$	N		AVERAGE N
			grams	mgm.	
1	Normal	None	3.5168		
2	Normal	None	3.5364		3.5266
3	Normal	0.189	3.5978		
4	Normal	0.189	3.6700		3.6339
5	Normal	0.378	3.8444		
6	Normal	0.378	3.9452		3.8948
7	Normal	0.756	4.2728		
8	Normal	0.756	4.3736		4.3232
9	Normal	1.512	4.7432		
10	Normal	1.512	4.7824		4.7628
11	Normal	6.048	1.1340		
12	Normal	6.048	1.0612		1.0976

plied up to a treatment of 6 per cent. Somewhere between 1.512 per cent and 6.048 per cent occurred the toxic point for this salt on this particular soil. Unfortunately, the exact point was not ascertained. It is of considerable interest to note; however, the undoubtedly toxicity to nitrification indicated by the application of 6.048 per cent. Evidently calcium carbonate in small or reasonable amounts is beneficial to nitrification, but when the addition becomes excessive there is a depression in the activities of this group of or-

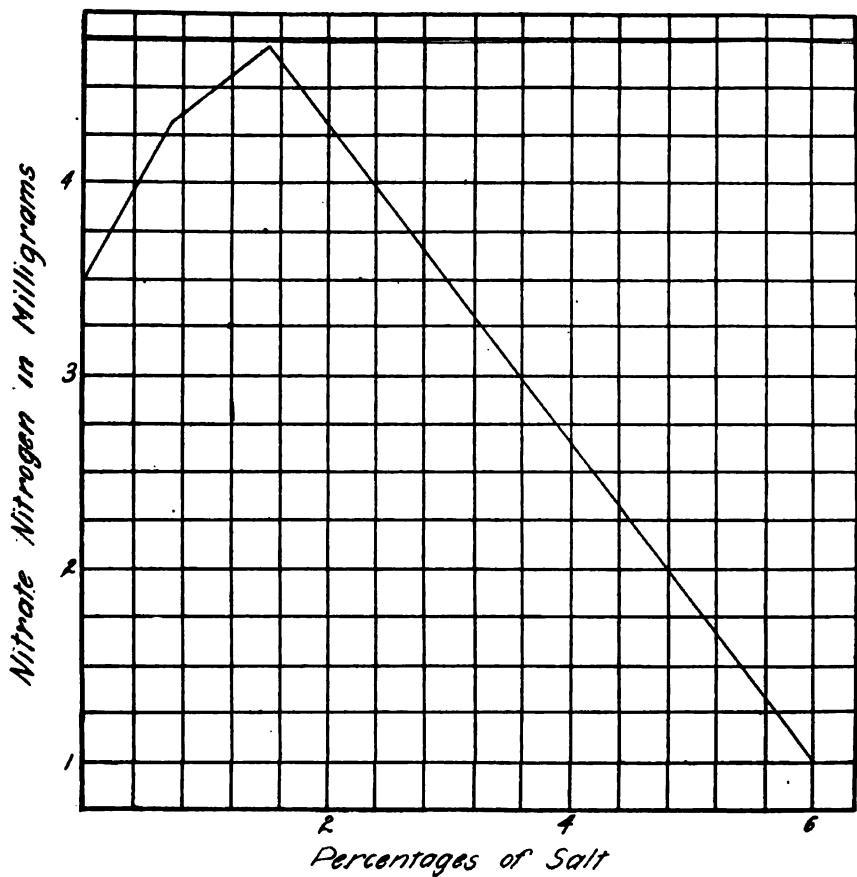


FIG. 4. EFFECTS OF CaCO_3 ON NITRIFICATION IN NORMAL SOIL

ganisms. This depression may not occur in other soils, but these results on one particular soil indicate that calcium carbonate may be applied to soils in too large amounts.

Series V. The effects of NaHCO_3 on nitrification in alkali soil

This series deals with the effects of sodium bicarbonate on nitrification in an alkali soil. The treatment of the soil and the results appear in table 5 and figure 5. Examining the table, it is seen that the NaHCO_3 additions up to and beyond 0.10 per cent stimulated considerably the nitrification process in this soil. Beyond 0.30 per cent, however, a distinct depression occurred which increased with increasing additions.

When calcium sulfate was added with the sodium bicarbonate, all toxic effects of the additions were removed, as is indicated by the results of the last two tests in the table.

TABLE 5
Effects of NaHCO₃ on nitrification in alkali soil

NUMBER	SOIL	NaHCO ₃ grams	N	AVERAGE N mgm.
			mgm.	
1	Alkali	None	4.6172	
2	Alkali	None	4.5416	4.5794
3	Alkali	0.05	4.8740	
4	Alkali	0.05	4.7650	4.8190
5	Alkali	0.10	5.4740	
6	Alkali	0.10	5.3228	5.3984
7	Alkali	0.30	3.8864	
8	Alkali	0.30	4.2148	4.0506
9	Alkali	0.50	3.8360	
10	Alkali	0.50	3.9116	3.8738
11	Alkali	1.00	3.7604	
12	Alkali	1.00	3.1808	3.4706
13*	Alkali	1.00	4.9448	
14*	Alkali	1.00	4.8440	4.8944

* Plus 1.6197 grams CaSO₄.

Sodium bicarbonate appears, from these results, to have a stimulative effect on nitrification when added to soil to 0.10 to 0.30 per cent, but it becomes toxic to the process when applied at the rate of 0.30 per cent and beyond, the toxicity increasing with the addition.

(With CaSO₄ ---)

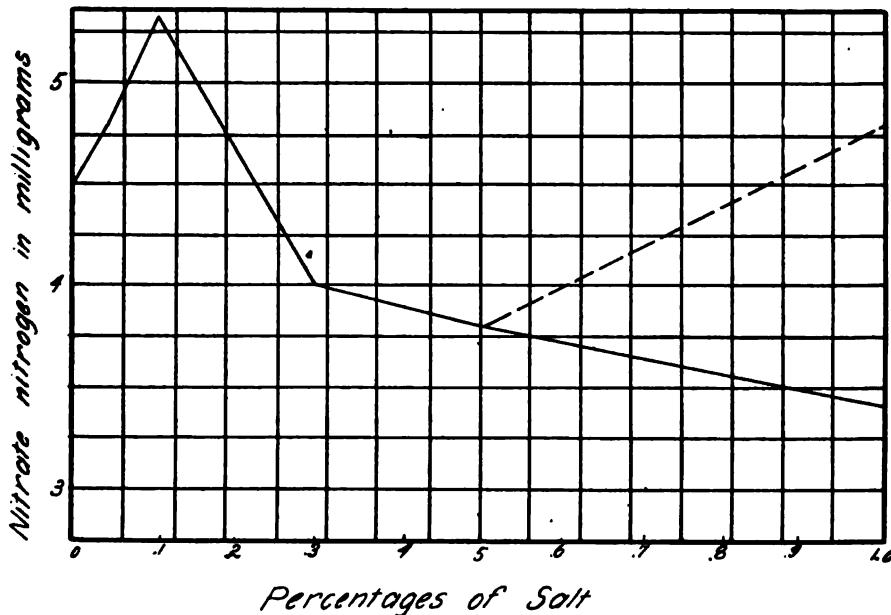


FIG. 5. EFFECTS OF NaHCO₃ ON NITRIFICATION IN ALKALI SOIL

When calcium sulfate is added with the sodium bicarbonate in the proper amount to react with this latter salt, all toxic effect is removed and there is apparently no influence from the addition, except perhaps a slight increase in the nitrification process.

Series VI. The effects of Na_2CO_3 on nitrification in alkali soil

The arrangement of this series was very similar to the preceding. It is shown in table 6 and figure 6 together with the results.

TABLE 6
Effects of Na_2CO_3 on nitrification in alkali soil

NUMBER	SOIL	Na_2CO_3 grams	N mgm.	AVERAGE N
				mgm.
1	Alkali	None	2.1476	
2	Alkali	None	1.7948	1.9712
3	Alkali	0.05	3.1528	
4	Alkali	0.05	1.8480	2.5004
5	Alkali	0.10	1.6436	
6	Alkali	0.10	2.5256	2.0846
7	Alkali	0.30	2.2484	
8	Alkali	0.30	1.6184	1.9334
9	Alkali	0.50	1.9936	
10	Alkali	0.50	1.5176	1.7556
11	Alkali	1.00	1.2656	
12	Alkali	1.00	1.7416	1.5036
13*	Alkali	1.00	2.1252	
14*	Alkali	1.00	1.8480	1.9866

* Plus 1.30 grams CaSO_4 .

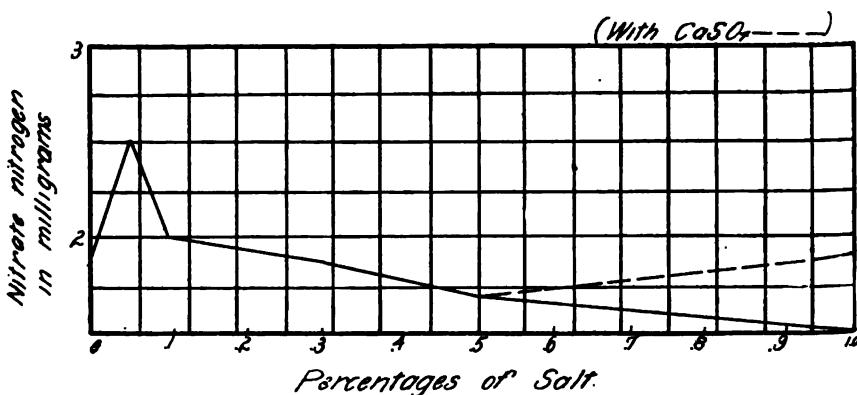


FIG. 6. EFFECTS OF Na_2CO_3 ON NITRIFICATION IN ALKALI SOIL

The application of the sodium carbonate at the rate of 0.05 per cent gave a decided stimulation in nitrification. The 0.10 per cent addition, likewise, gave a stimulation, but it was not large. The larger additions brought about

depressions in the process which became greater as the size of the application increased. When calcium sulfate was added in the proper amount to react with the sodium carbonate there was no effect whatever from the addition. This result is in accord with that of the preceding series with the bicarbonate of sodium and the calcium sulfate.

The concentration at which the sodium carbonate became toxic to the nitrifiers in this soil was much greater than that noted by Lipman. He found toxicity at 0.025 per cent, but stimulation occurred in this work up to 0.10 per cent. His results with Na_2CO_3 , however, were not considered satisfactory, as there was much mold growth on the soils which may have used up the nitrates produced, and losses may have occurred in the several filtrations through bone black which were necessary to the determination. However, the variation from Lipman's results may be due, as was concluded in the preceding series, to differences in the soil used. It is evident, however, that sodium carbonate, like the other salts tested, stimulated nitrification when used in small amounts, but depressed the process considerably when large applications were made. When calcium sulfate is used with the sodium carbonate, there is no effect on the nitrifying bacteria.

Series VII. The effects of CaSO_4 on nitrification in alkali soil

The results in the preceding tables indicated that calcium sulfate applied with sodium carbonate and sodium bicarbonate neutralized the toxic effects

TABLE 7
Effects of CaSO_4 on nitrification in alkali soil

NUMBER	SOIL	CaSO_4	N	AVERAGE N
			grams	mgm.
1	Alkali	None	4.1804	
2	Alkali	None	4.3120	4.2462
3	Alkali	0.022	4.4156	
4	Alkali	0.022	4.4436	4.4298
5	Alkali	0.033	4.4436	
6	Alkali	0.033	4.5752	4.4894
7	Alkali	0.044	4.5472	
8	Alkali	0.044	4.6284	4.5878
9	Alkali	0.088	4.4156	
10	Alkali	0.088	4.3120	4.3638
11	Alkali	0.176	4.1664	
12	Alkali	0.176	4.3904	4.2784

of these salts. It seemed advisable, therefore, to ascertain the influence of the CaSO_4 when applied alone to the alkali soil. The treatments used and the results secured appear in table 7 and figure 7.

It is apparent upon an examination of this table that the CaSO_4 had little effect on nitrification. There seemed to be a slight stimulation with the

smaller additions, but the differences were not large. With the larger amounts, no increase was secured. The applications were not increased beyond 0.176 per cent, so it is quite possible that further additions might have brought about an increase in nitrification. However, in the absence of sufficient data, general conclusions cannot be drawn. The differences secured seem too small to conclude a stimulation of the process, but the results may be considered to indicate the possibility of such a stimulation at least.

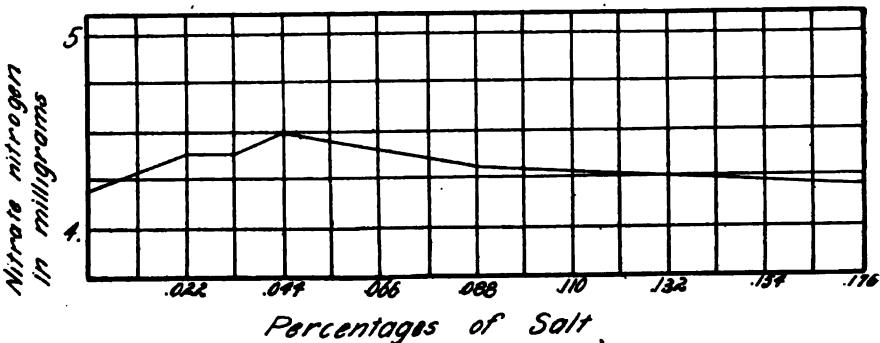


FIG. 7. EFFECTS OF CaSO_4 ON NITRIFICATION IN ALKALI SOIL

Series VIII. The effects of CaCO_3 on nitrification in alkali soil

When calcium carbonate was applied to the normal soil used in this work, a distinct stimulation of nitrification occurred up to 6.0 per cent. At that point, however, the activities of the nitrifiers were quite considerably depressed. It seemed desirable, therefore, to test the effects of that salt on nitrification in the alkali soil.

This series was arranged in a similar manner to series IV except that the alkali soil was used. The arrangement of the series and the results are given in table 8 and figure 8.

The small amounts of the CaCO_3 did not seem to influence nitrification, but when 0.378 per cent was used an increase in the process was noted. Further gains occurred with the larger applications up through 1.512 per cent. When 6.046 per cent was added, however, a distinct depression in nitrification occurred. Evidently the maximum increase in the process through the use of CaCO_3 on this soil occurred at 1.512 per cent, or between that amount and 6.048 per cent.

The absence of tests using intermediate amounts prevents any definite point being established. It is evident, however, that just as was the case in the tests with the normal soil, CaCO_3 when applied in small amounts to this alkali soil increased nitrification, but when 6.048 per cent was used, a decided depression in the process occurred. Such an amount of CaCO_3 is rarely found in soil and hence, even if an injurious effect were found to be quite general (with many soils), which is extremely unlikely, no danger from a reasonable use of lime on soils need be apprehended.

TABLE 8
Effects of CaCO₃ on nitrification in alkali soil

NUMBER	SOIL	CaCO ₃ grams	N mgm.	AVERAGE N
				mgm.
1	Alkali	None	2.1586	
2	Alkali	None	1.8900	2.0243
3	Alkali	0.189	2.0188	
4	Alkali	0.189	2.0440	2.0314
5	Alkali	0.378	2.1700	
6	Alkali	0.378	2.2960	2.2330
7	Alkali	0.756	2.3960	
8	Alkali	0.756	2.4976	2.4468
9	Alkali	1.512	2.7134	
10	Alkali	1.512	2.9026	2.8080
11	Alkali	6.048	1.0108	
12	Alkali	6.048	0.7840	0.8974

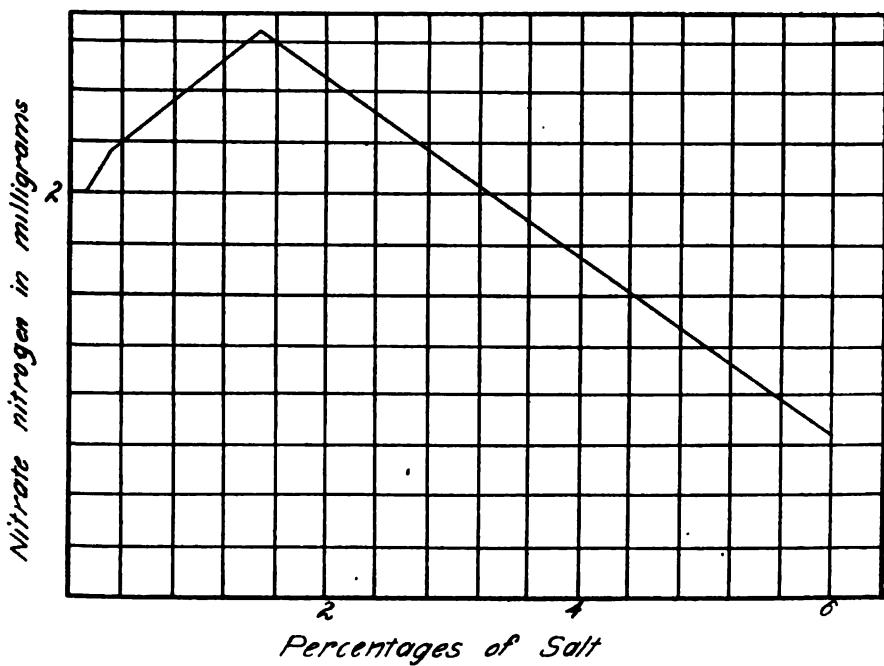


FIG. 8. EFFECTS OF CaCO₃ ON NITRIFICATION IN ALKALI SOIL

Series IX. The effects of NaHCO₃, Na₂CO₃ and CaSO₄ on nitrification in alkali soil

This series was planned to test the effect of the carbonate and bicarbonate of sodium on nitrification in alkali soil in the presence of calcium carbonate and with and without the addition of sufficient calcium sulfate to react with the carbonate used. The arrangement of the tests and the results secured are given in table 9 and figure 9.

TABLE 9
Effects of NaHCO₃, Na₂CO₃, and CaSO₄ on nitrification in the presence of CaCO₃ in alkali soil

NUMBER	ADDITION	N	AVERAGE N
		mgm.	mgm.
1	Nothing.....	2.3296	
2	Nothing.....	2.5312	2.4304
3	0.536 gram CaCO ₃	2.9960	
4	0.536 gram CaCO ₃	4.1440	3.5700
5	0.536 gram CaCO ₃ + 1.00 gram NaHCO ₃	2.6852	
6	0.536 gram CaCO ₃ + 1.00 gram NaHCO ₃	2.5760	2.6306
7	0.536 gram CaCO ₃ + 0.044 gram NaHCO ₃ + 0.072 gram CaSO ₄	2.4640	
8	0.536 gram CaCO ₃ + 0.044 gram NaHCO ₃ + 0.072 gram CaSO ₄	2.9688	2.7164
9	0.536 gram CaCO ₃ + 0.044 gram NaHCO ₃	Lost	
10	0.536 gram CaCO ₃ + 0.044 gram NaHCO ₃	3.0016	3.0016
11	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃	2.7104	
12	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃	2.7552	2.7328
13	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃ + 0.072 gram CaSO ₄	3.0912	
14	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃ + 0.072 gram CaSO ₄	4.2928	3.6920
15	0.536 gram CaCO ₃ + 1.00 gram Na ₂ CO ₃	2.8224	
16	0.536 gram CaCO ₃ + 1.00 gram Na ₂ CO ₃	Lost	2.8224
17	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃ + 0.044 gram NaHCO ₃	2.5760	
18	0.536 gram CaCO ₃ + 0.056 gram Na ₂ CO ₃ + 0.044 gram NaHCO ₃	3.4272	3.0016

On examining this table it is apparent that calcium carbonate increased the nitrification process to a considerable extent. This is in accord with the tests previously discussed with calcium carbonate. The sodium carbonate and the bicarbonate both depressed considerably the activities of the nitrifiers over calcium carbonate alone, the larger application bringing about a greater depression in the case of the bicarbonate, but practically the same results being secured with both amounts in the case of the carbonate.

Some differences are evident here from the results secured with the use of the two salts without calcium carbonate. In those cases the small amount

of the carbonates increased nitrification slightly, but in the presence of CaCO_3 in this series, a depression occurred. This is an interesting difference and points to the fact that amounts of these salts which alone are non-toxic may become toxic when present with CaCO_3 .

When calcium sulfate was used with the smaller amount of the sodium carbonate, the injurious effect of the carbonate was neutralized and no influence on nitrification is noted. This is in accord with the results secured with the two salts in the absence of CaCO_3 . When the bicarbonate is considered, however, the use of the calcium sulfate with the bicarbonate depressed the process more than was the case with the bicarbonate alone. This is exactly opposite from the results secured in the previous series in the ab-

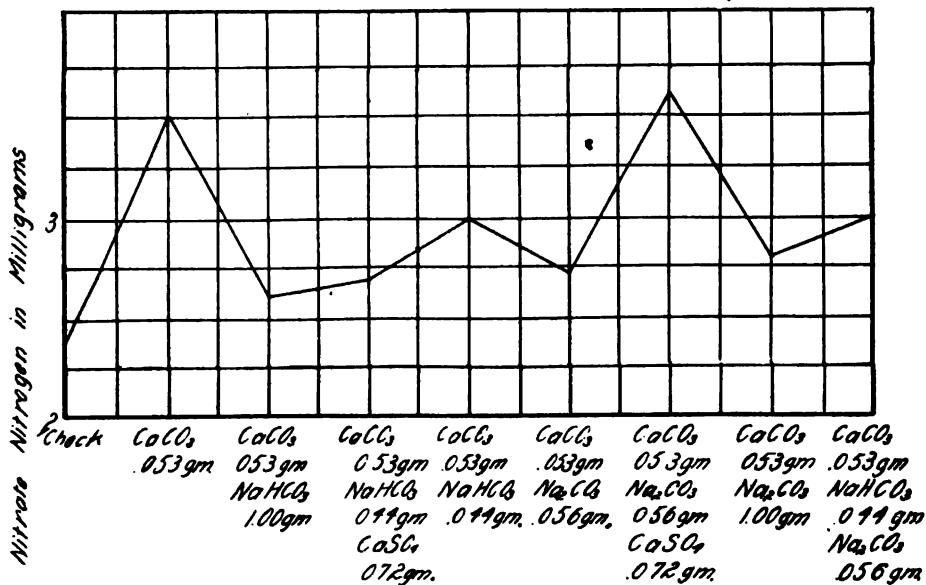


FIG. 9. EFFECTS OF CaCO_3 AND NaHCO_3 , Na_2CO_3 AND CaSO_4 IN THE PRESENCE OF CaCO_3 ON NITRIFICATION IN ALKALI SOIL

sence of CaCO_3 . Only one determination was made with the use of the bicarbonate alone, and hence that result may not be absolutely accurate, but the differences secured are definite enough so that it must be concluded that in the presence of CaCO_3 calcium sulfate does not neutralize the toxic effect of sodium bicarbonate.

When the bicarbonate and carbonate of sodium were applied together, the depression in nitrification was less than with the sodium carbonate alone and about the same as that from the bicarbonate alone.

These results, as a whole, show distinctly the toxic effects of the carbonate and bicarbonate of sodium on nitrification in this soil in the presence of CaCO_3 . Furthermore, smaller amounts became toxic than in the same soil

in the absence of CaCO_3 . CaSO_4 prevents the toxic action of sodium carbonate, but it does not seem to be similarly efficient with the bicarbonate in the presence of CaCO_3 .

GREENHOUSE EXPERIMENTS

Comparative experiments similar to the above were carried out in the greenhouse, nitrification tests being run on both the normal and alkali soils, after beans had been grown and harvested. In these pot experiments 10 pounds of soil were used and the treatment of the normal soil was calculated to imitate alkali-soil conditions. The amounts of the various alkali salts added singly or in combination to the normal soil were the same as those found

TABLE 10
Effects of NaCl , Na_2SO_4 , MgSO_4 and CaSO_4 on nitrification in normal soil

NUM- BER	TREATMENT	FIRST SAMPLING		SECOND SAMPLING		THIRD SAMPLING	
		Average N	Average N for pots	Average N	Average N for pots	Average N	Average N for pots
		mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
1	None.....	6.6570		6.6010		6.9060	
2	None.....	6.2748	6.4659	6.2916	6.4463	6.7088	6.8074
3	0.005 per cent NaCl	6.1562		6.5464		7.0280	
4	0.005 per cent NaCl	6.0088	6.0825	6.5874	6.5664	7.0168	7.0224
5	0.518 per cent Na_2SO_4	6.5562		6.5996		6.3168	
6	0.518 per cent Na_2SO_4	5.9066	6.2314	6.3476	6.4736	6.4932	6.4050
7	0.023 per cent MgSO_4	7.4466		5.8660		6.8368	
8	0.023 per cent MgSO_4	6.5940	7.0203	6.4381	6.1520	6.9622	6.8995
9	0.189 per cent CaCO_3	6.4932		6.6280		7.7420	
10	0.189 per cent CaCO_3	7.3234	7.1583	6.7340	6.6810	6.6870	7.2145
11	All four salts.....	6.4288		6.4932		6.8152	
12	All four salts.....	6.5940	6.5114	6.1278	6.3105	7.0158	6.9155

by analysis in the alkali soil. The alkali soil received additions of calcium carbonate and of sodium carbonate and bicarbonate with and without additions of calcium sulfate.

All the treatments were in duplicate, and the arrangement of the pots is shown in tables 10 and 11.

The pots were weighed at the time of filling and brought up to the optimum moisture content, which was 20.0 per cent for the normal soil and 20.8 per cent for the alkali soil. The moisture content was kept constant throughout the experiment by adding water to weight. This water was added by means of a tube at the bottom of the pots, in order to prevent puddling. The experiment was started on November 12, 1915, and beans seeded in all the pots in the attempt to ascertain whether the effects of the alkali salts would be the same on the crop grown as on the nitrifying organisms. After the crop was removed, samples were drawn and tested for nitrifying power on February

23, March 10 and March 20 for the normal soils, and on February 6, February 25, March 12 and March 22 for the alkali soils. The procedure in these tests was the same as that already outlined, except that fresh soil was used, no soil infusion was added, and of course, no salts were used, the idea being to determine the nitrifying power of the soils receiving the various salts *after* a crop had been grown and *after* the salts had been present in the soil for a comparatively long period.

The results of the tests of the normal soils are given in table 10 and figures 10 and 11, and of the alkali soils in table 11 and figures 12 and 13.

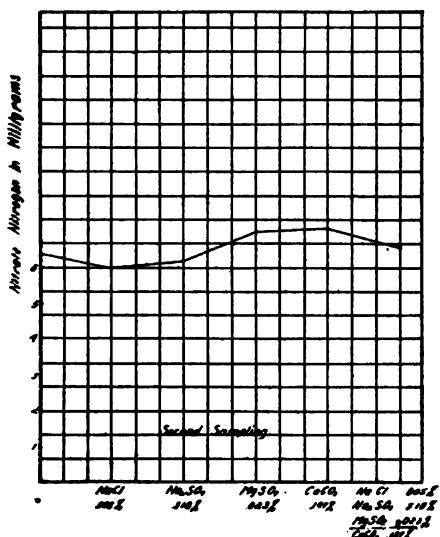


FIG. 10

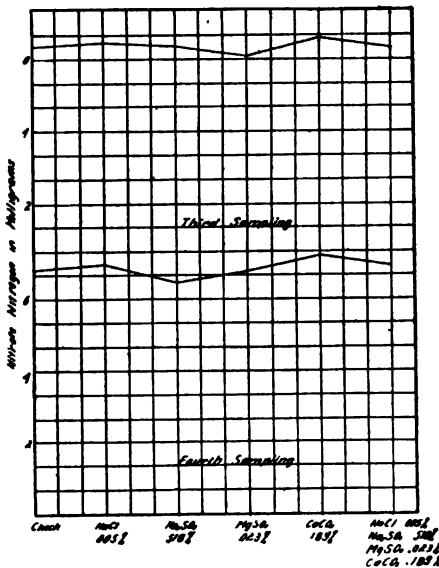


FIG. 11

FIG. 10. EFFECTS OF NaCl , Na_2SO_4 , MgSO_4 AND CaCO_3 ON NITRIFICATION IN NORMAL SOIL—SECOND SAMPLING (POT EXPERIMENTS)

FIG. 11. EFFECTS OF NaCl , Na_2SO_4 , MgSO_4 , AND CaCO_3 ON NITRIFICATION IN NORMAL SOIL—THIRD AND FOURTH SAMPLINGS (POT EXPERIMENTS)

Normal soil tests

Considering the results as a whole, it is apparent from table 10 that sodium chloride gave a very slight increase in nitrification. This increase was quite noticeable at the last two samplings. This effect of the use of 0.005 per cent of NaCl checks exactly with the results secured in the laboratory test with the use of NaCl as shown in table 1. Evidently the stimulative effect of the NaCl on nitrification when amounts not exceeding 0.005 per cent were used continues for some time after the application is made, if these results in the pots are considered sufficiently definite to warrant conclusions. At any rate, no toxic effect is seen. Na_2SO_4 gave a slight toxic effect on the nitrifying power of the soil, as shown at the first and last samplings. A very slight increase

TABLE II
Effects of CaCO_3 , NaHCO_3 , Na_2CO_3 and CaSO_4 on nitrification in alkali soil

NUMBER	TREATMENT	FIRST SAMPLING		SECOND SAMPLING		THIRD SAMPLING		FOURTH SAMPLING	
		Average N mgm.	Average N for pots mgm.						
1	None.....	4.1370	4.6844	4.5150	5.4955				
2	None.....	4.0614	4.0992	4.8454	4.7149	4.7264	4.6207	5.2106	5.3531
3	0.189 per cent CaCO_3	5.4642		4.9784		4.5262		5.4950	
4	0.189 per cent CaCO_3	5.6014	5.5328	4.6592	4.8183	4.5703	4.5482	5.4418	5.4684
5	0.378 per cent CaCO_3	Lost		4.7348		4.7454		5.8422	
6	0.378 per cent CaCO_3	5.5384	5.5384	5.8954	5.3151	5.1310	4.9382	5.3998	5.6210
7	0.109 per cent NaHCO_3	4.8174		5.0414		4.9560		5.9528	
8	0.109 per cent NaHCO_3	4.6601	4.7384	5.2696	5.0435	4.4716	4.7138	4.9518	5.4523
9	0.218 per cent Na_2CO_3	4.9518		4.9261		4.5402		5.9010	
10	0.218 per cent Na_2CO_3	4.8622	4.9070	6.1726	5.5493	4.8342	4.6872	5.7512	5.8261
11*	0.109 per cent NaHCO_3	4.9644		6.0970		4.8384		5.7512	
12*	0.109 per cent NaHCO_3	4.7710	4.8677	6.0088	6.0529	4.8230	4.8307	6.2216	5.9864
13†	0.218 per cent Na_2CO_3	Lost		5.6168		4.9560		7.0424	
14†	0.218 per cent Na_2CO_3	5.5916	5.5916	6.5170	6.0669	5.1310	5.0440	6.5324	6.7874

* Plus 0.177 per cent CaSO_4 .

† Plus 0.261 per cent CaSO_4 .

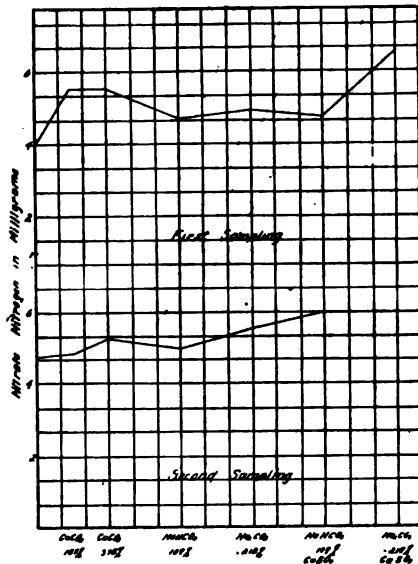


FIG. 12

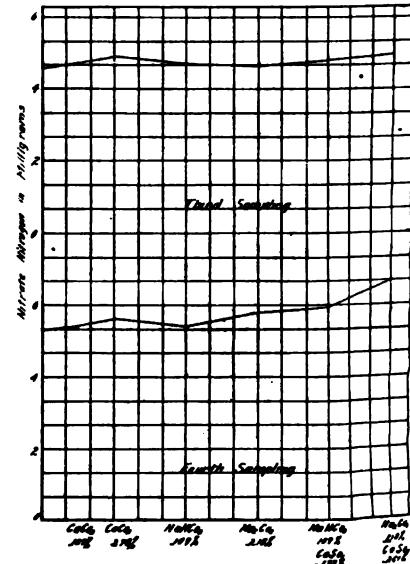


FIG. 13

FIG. 12. EFFECTS OF CaCO_3 , NaHCO_3 , Na_2CO_3 AND CaSO_4 ON NITRIFICATION IN ALKALI SOIL—FIRST AND SECOND SAMPLINGS (POT EXPERIMENTS)

FIG. 13. EFFECTS OF CaCO_3 , NaHCO_3 , Na_2CO_3 , AND CaSO_4 ON NITRIFICATION IN ALKALI SOIL—THIRD AND FOURTH SAMPLINGS (POT EXPERIMENTS)

occurred at the second sampling, but the difference was too small to be considered significant. The laboratory tests showed a stimulative effect of 0.518 per cent of Na_2SO_4 on nitrification but the effect evidently disappears after the salt has been in the soil for several months and a slight toxic effect is found from this amount of this salt. MgSO_4 gave a beneficial effect on nitrification at all samplings except the second, and this is in accord with the results secured in series III, where 0.022 per cent of MgSO_4 gave a distinct increase in the process. The effect was not so great here and evidently the beneficial influence of the salt gradually decreased as the time since application increased. After a still longer period it is possible that there would have been no effect whatever or the opposite influence from this salt, just as was noted with the Na_2SO_4 .

CaCO_3 in all cases gave a pronounced increase in nitrification and this result checks with that secured in the laboratory test. The effects of this salt evidently persist for a longer period after it is applied to the soil than is the case with the other salts just mentioned. This salt is so essential in soils for the process of nitrification to occur to the best advantage that its effect in increasing the process is easily understandable.

When all four salts were applied together there was no effect on nitrification. The beneficial effect of the calcium carbonate just noted was not apparent in the presence of the other salts. The slight toxic effect from the Na_2SO_4 noted when it was used alone evidently increases in the presence of calcium carbonate and this is offset by the beneficial influence of the CaCO_3 . Harris (4) found this fact to be true in his tests of the alkali salts on crop growth. It also forms a basis for the result secured in series VIII showing the toxic effect of CaCO_3 when applied in larger quantities to the alkali soil.

Alkali soil tests

On examining the results of the nitrification tests in the soils from the alkali pots it is seen that CaCO_3 in both amounts used increased the nitrifying power of the soil. The larger amount gave the larger increase in all cases. The gains were generally quite considerable. Likewise, the NaHCO_3 gave quite considerable increases in the nitrifying power of the soils. These results are in agreement with the laboratory tests in series V which showed decided benefit to nitrification from the use of 0.10 per cent NaHCO_3 . That series showed larger amounts than this to be toxic to the nitrifiers and experiments of others have shown that larger amounts are injurious, hence it is evident that this alkali soil does not contain any considerable amount of NaHCO_3 . With Na_2CO_3 , very similar results were secured as with the bicarbonate. Distinct increases in nitrifying power were noted at every sampling. These results check fairly accurately with those secured in series VI which showed a stimulation in nitrification from the application of 0.10 per cent and a depression at 0.30 per cent. It is quite possible that the turning point occurred in that case at about 0.30 per cent and that 0.20 per

cent would have shown an increase. Larger amounts of this salt, like the bicarbonate, depress nitrification, and hence it would seem that this alkali soil is not extremely high in this salt. When calcium sulfate was used with the sodium salts, in every case, with both salts there was an increased stimulation in nitrification. The calcium sulfate alone in the laboratory tests in series VII showed no effect on nitrification in the alkali soil, so the effect here is evidently due to the combinations with the sodium salts. Series IX showed a slight increase in nitrification when CaSO_4 was applied with Na_2CO_3 in the presence of CaCO_3 , and that result may serve to verify the present. NaHCO_3 in that series did not produce this effect. It is apparent from these results that the nitrification process in this alkali soil was not low as a result of any excess of sodium carbonate or bicarbonate. Calcium carbonate in large amounts likewise did not depress nitrification, so the alkali condition is evidently not due to excess of this salt in the alkali soil. The laboratory tests and the tests in the greenhouse soil several months after treatment were in excellent agreement.

Crop experiment

As has been mentioned, beans were seeded in both the normal and alkali soils in the greenhouse tests. Four separate plantings were made in the

TABLE 12
Crop results of pots containing normal soils

POT NUM-BER	TREATMENT	GREEN WEIGHT OF CROP	AVERAGE
			GRAMS
1	None.....	33	
2	None.....	39	36.0
3	NaCl —0.005 per cent.....	41	
4	NaCl —0.005 per cent.....	39	40.0
5	Na_2SO_4 —0.518 per cent.....	31	
6	Na_2SO_4 —0.518 per cent.....	31	31.0
7	MgSO_4 —0.023 per cent.....	49	
8	MgSO_4 —0.023 per cent.....	46	47.5
9	CaCO_3 —0.189 per cent.....	42	
10	CaCO_3 —0.189 per cent.....	38	40.0
11	All four salts.....	35	
12	All four salts.....	34	34.5

alkali soil but the seed refused to germinate. Barley was then seeded, but grew only in the pot receiving CaCO_3 , and the per cent of germination was very small. No crop results in the alkali soil were secured, therefore.

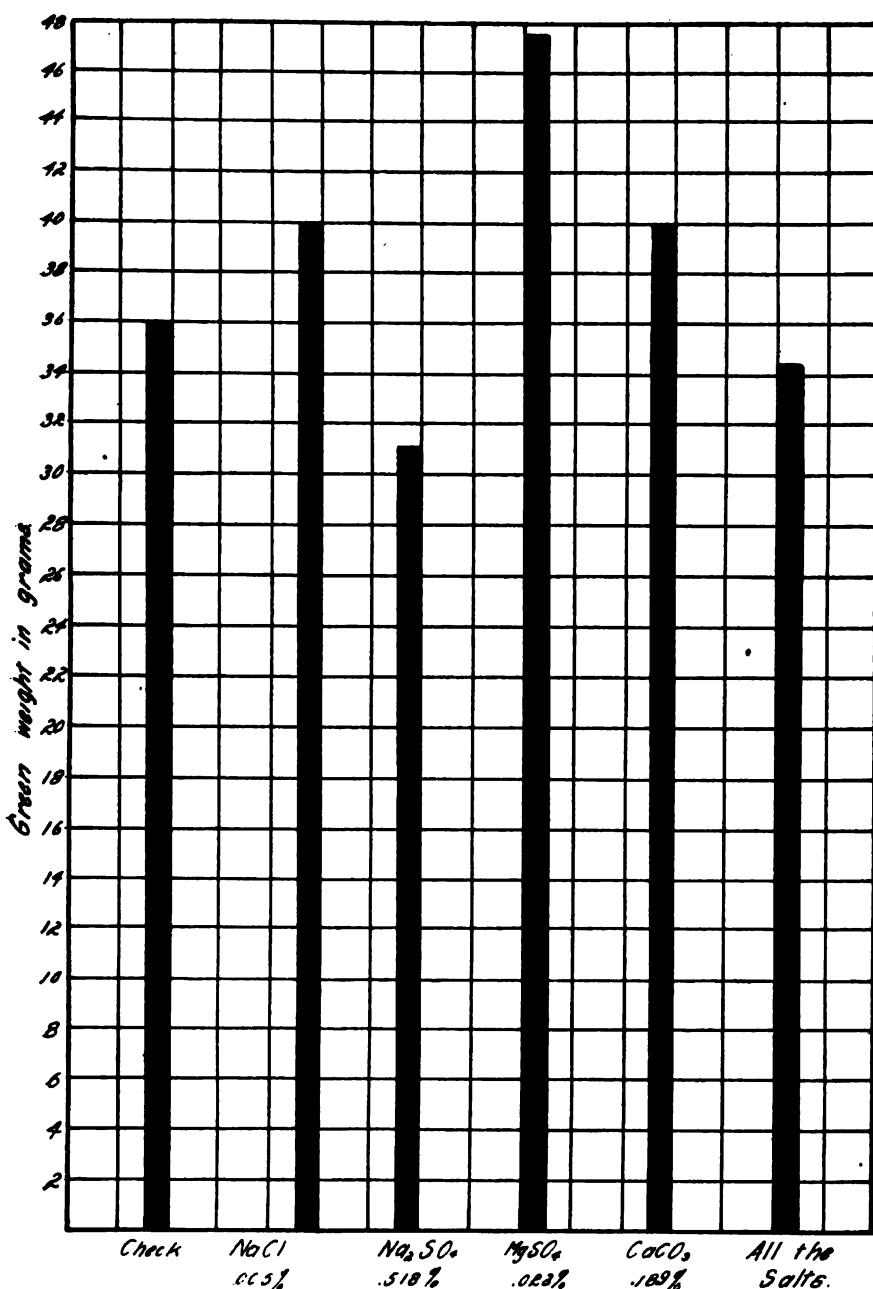


FIG. 14. EFFECTS UPON BEANS GROWN IN NORMAL SOIL WITH THE ADDITION OF VARIOUS
ALKALI SALTS

In the normal soil the bean crop was harvested on February 4, and the green weight secured. The results in table 12 and figure 14 show the crop yields. The NaCl gave an increase in the crop, as did the MgSO₄ and CaCO₃; the Na₂SO₄ depressed the yield slightly and the combination of all four salts gave no effect on the crop. These results check quite accurately with the results of the nitrification tests in the greenhouse soils and, except for the Na₂SO₄, with the results in the laboratory. Na₂SO₄ in these latter tests increased nitrification. The amounts of NaCl, MgSO₄ and CaCO₃ used were beneficial, both to crop growth and to nitrification, while the Na₂SO₄ was injurious, except in the laboratory test. All the salts together had no effect. Lipman's (8, 9, 10) conclusion that nitrification tests and crop effects agree, in the case of alkali salt studies is thus confirmed. These crop results agree very well with those of Harris (4) already referred to.

SUMMARY AND CONCLUSIONS

These studies on the effects of alkali salts on nitrification in a normal soil from Wyoming, and on comparative nitrification in normal soil variously treated and in alkali soil, untreated and treated, gave results which, while perhaps not conclusive, are at least indicative of certain relations in the field. These indications may be summarized thus:

1. Nitrification in normal soil is stimulated by small amounts of NaCl, Na₂SO₄ and MgSO₄, and large amounts of CaCO₃. These salts become toxic, however, at certain points, which undoubtedly vary in different soils. With this soil in laboratory tests the toxic point was 0.02 per cent NaCl, 2.00 per cent Na₂SO₄ and between 1.5 and 6.00 per cent CaCO₃. The toxic point for MgSO₄ was not determined.
2. Nitrification in alkali soil was increased by small amounts of NaHCO₃, Na₂CO₃ and CaCO₃. Calcium sulfate had no effect. These salts became toxic in this soil at 0.30 per cent for both the sodium carbonate and bicarbonate and at 6.0 per cent for the CaCO₃. The addition of CaSO₄ with the sodium carbonate and bicarbonate, in the proper amount to react with them, prevented any toxic effect from the largest amount used.
3. The tests in the greenhouse soils checked very closely with the laboratory studies in the case of the alkali soils. In the normal soils the agreement was likewise good, except in the case of Na₂SO₄. That salt became toxic according to these tests at a concentration of 0.5 per cent. This is a very much lower toxic point than was noted above but nearer that found by others.
4. The effect on the crop grown in normal soil of the alkali salts, with the exception of the Na₂SO₄, were very similar to the effects on nitrification in both laboratory and greenhouse tests. Increases were secured with NaCl, MgSO₄ and CaCO₃, but Na₂SO₄ caused a depression in crop and in nitrification in the greenhouse soils. All the salts together had no effect. In general, it seems that nitrification and crops are very similarly affected by alkali salts.

5. Crops refused to grow in the alkali soil, but the injurious factor was evidently not an excess of NaHCO_3 or Na_2CO_3 , as additions of these salts increased nitrification in the soil. The injurious factor was likewise evidently not CaCO_3 , for that compound stimulated nitrification in the alkali soil.

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THE DIVERGENT EFFECTS OF LIME AND MAGNESIA UPON THE CONSERVATION OF SOIL SULFUR

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Until recently scant consideration was accorded the element sulfur in the study of soil fertility. Inaccurate methods of ash analysis, which failed to show the entire amounts assimilated by plants, were probably in a large measure responsible for failure to attribute to sulfur an important rôle in the study of plant needs. It was therefore natural that slight attention should be paid to the form or extent of the element as it occurs in soil. But within the last several years particular attention has been given the subject from the several viewpoints of plant-ash composition, conservation of soil supply, and the amount derived from rainfall. The relationship between soil sulfur and biological activities also has been studied. The Wisconsin, Kentucky, Rothamsted, Ohio, Nebraska, and Iowa stations have contributed to the literature upon the several phases of the subject.

The results submitted in this paper were not obtained from a study which had its inception in a consideration of the sulfur problem. However, the findings as to the behavior of the element during the first two annual periods have been exceedingly striking. It is not intended at this time to offer a complete study or a full array of data obtained, nor of conclusions suggested thereby, but to submit the findings in the way of a preliminary report for the benefit of those who are working upon, or who are particularly interested in, the sulfur problem as related to soil fertility.

Before offering the data obtained, it is essential that a brief description of the experimental equipment and methods followed be given. A mellow sandy loam from the second bench of the Tennessee River at Knoxville was used. One hundred pounds of soil, on the moisture-free basis, were used for each treatment. The lysimeter equipment used in the work is illustrated in plate 1 (figs. 1 and 2). The soil leaching studies now being carried out in the equipment were intended to supplement the work of this station upon the problem of absorption of the several forms of lime and magnesia by soil under field conditions. In the preliminary work, reported upon in part in Tennessee Agricultural Experiment Station Bulletin 107, both 2- and 8-ton applications of lime per acre, or equivalent amounts of the several forms of the earthy alkalies, were made. In the supplementary work here considered, the treatments were based upon chemical equivalences of 8, 32 and 100 tons of CaO per acre. The

excessive applications were made with the idea of affording an opportunity for a study of fundamentals, rather than delimiting the work to bounds which would yield data susceptible of only practical application. The following seven alkali-earth materials were used: burnt lime, burnt magnesia, precipitated calcium carbonate, precipitated magnesium carbonate, 100-mesh limestone, 100-mesh dolomite, and 100-mesh magnesite. Each treatment was thoroughly mixed with moist soil in good, workable condition and placed in a galvanized iron lysimeter containing a sand filter bed, and having a block tin drainage tube. In a second set, placed simultaneously, the foregoing 21 treatments were duplicated as to surface soil, but in this set one foot of clay subsoil was placed between each sand filter and the overlying surface soil. Two additional tanks also were used as separate containers for the relatively inactive natural silicates, wollastonite, and serpentine. Blanks of both depths also were later included.

The successive leachings, derived from natural rainfall only, from July, 1914, to July, 1916, were conveyed to the laboratory, where their respective alkalinites were immediately determined. Aliquots were then preserved in acid solution and the composites for each year were subjected to complete analysis.

In the fifth and sixth columns of table I are given the total alkalinites of the several yearly leachings, expressed as cubic centimeters of half normality. In the seventh and eighth columns are given the amounts of SO₃ per acre for the two respective annual periods. A number of tests were made upon the several solutions in order to determine the form of the sulfur present. It appeared that all of the sulfur leached was in the form of sulfates.

A study of table 1 shows certain positive and consistent differences, particularly as to the estoppage effected by the subsoil. During the first year the loss of sulfur was very much heavier from the tanks containing only surface soil than from the tanks which contained subsoil also. As a rule the same holds for the second year; but a marked exception is to be observed in the instances where the oxide and precipitated carbonate of magnesium were applied. The averages of the total amounts of SO₃ leached from all the tanks receiving the several carbonates are 472 pounds and 221 pounds per acre, respectively, for the years 1914-1915 and 1915-1916, in the case of the tanks having no subsoil, as compared with 31.1 pounds and 114.8 pounds per acre, respectively, for the identically treated tanks during the same two years where the surface soil was underlaid with one foot of clay subsoil. The material increase in the average of SO₃ leached through the subsoil during the second year is due mainly to the fact that during that period large amounts of sulfur began to leach from the tanks containing the treatments of oxide and precipitated carbonate of magnesium, and from these tanks only. Analyses of the leachings established the fact that the downward movement of sulfur and that of magnesium were parallel. That is, where no subsoil was placed, large quantities of both sulfur and magnesium leached out of the surface soil

TABLE 1.

Alkalinity of leachings and amount of sulfur trioxide leached in two years' tests

TANK NO.	TREATMENT Substance applied	Rate- tons per acre	AMOUNT OF SUBSOIL	TOTAL ALKALINITY OF LEACHINGS— CUBIC CENTI- METERS N/2		SO ₃ LEACHED— POUNDS PER ACRE PER ANNUM	
				1914-15	1915-16	1914-15	1915-16
1	CaO.....	8	None	449	604	341.0	267.0
22	CaO.....	8	1 foot	102	174	32.9	68.2
8	CaO.....	32	None	1642	592	19.0	116.0
29	CaO.....	32	1 foot	114	98	25.0	21.6
15	CaO.....	100	None	4301	5011	67.0	15.0
36	CaO.....	100	1 foot	119	105	17.4	29.5
2	MgO.....	8	None	1024	937	469.0	141.0
23	MgO.....	8	1 foot	98	153	40.1	161.6
9	MgO.....	32	None	2572	4089	567.0	216.0
30	MgO.....	32	1 foot	65	476	18.0	488.7
16	MgO.....	100	None	1120	2392	581.0	291.0
37	MgO.....	100	1 foot	95	99	24.6	228.7
3	CaCO ₃	8	None	165	434	468.0	203.0
24	CaCO ₃	8	1 foot	105	140	34.9	58.4
10	CaCO ₃	32	None	279	442	448.0	187.0
31	CaCO ₃	32	1 foot	122	141	29.3	30.1
17	CaCO ₃	100	None	250	399	460.0	287.0
38	CaCO ₃	100	1 foot	98	133	27.0	124.0
4	MgCO ₃	8	None	2207	1356	528.0	170.0
25	MgCO ₃	8	1 foot	98	163	31.7	136.2
11	MgCO ₃	32	None	2365	3223	551.0	203.0
32	MgCO ₃	32	1 foot	170	451	54.9	593.6
18	MgCO ₃	100	None	2418	3833	618.0	364.0
39	MgCO ₃	100	1 foot	94	217	38.4	409.7
5	Ground limestone.....	8	None	379	479	540.0	214.0
26	Ground limestone.....	8	1 foot	89	128	19.5	29.1
12	Ground limestone.....	32	None	330	459	399.0	172.0
33	Ground limestone.....	32	1 foot	111	125	31.4	27.5
19	Ground limestone.....	100	None	392	484	402.0	215.0
40	Ground limestone.....	100	1 foot	164	161	40.4	87.2
6	Ground Dolomite.....	8	None	230	376	401.0	215.0
27	Ground Dolomite.....	8	1 foot	101	155	20.7	27.4
13	Ground Dolomite.....	32	None	311	548	402.0	238.0
34	Ground Dolomite.....	32	1 foot	118	122	38.8	77.7
20	Ground Dolomite.....	100	None	298	528	417.0	325.0
41	Ground Dolomite.....	100	1 foot	123	132	26.5	18.4
7	Ground Magnesite.....	8	None	489	679	512.0	171.0
28	Ground Magnesite.....	8	1 foot	118	158	30.6	36.0
14	Ground Magnesite.....	32	None	873	859	426.0	176.0
35	Ground Magnesite.....	32	1 foot	140	178	21.1	44.6
21	Ground Magnesite.....	100	None	433	785	509.0	181.0
42	Ground Magnesite.....	100	1 foot	131	160	22.0	21.8
43	Wallostonite.....	32	1 foot	177	170	35.3	26.2
44	Serpentine.....	32	1 foot	170	174	42.1	18.6

treated with the oxide and carbonate of magnesium during both years. However, although the subsoil arrested the passage of both magnesium and sulfur during the first year, it permitted the passage of both elements during the second year. The elimination of the results from the magnesium carbonate tanks materially alters the average of the subsoil tanks for the second annual period. If we average the leachings from the tanks containing treatments of the natural carbonates of both calcium and magnesium and also those containing precipitated carbonate of lime, the alkali earths in all of which are still in large measure retained by the subsoil, we find a loss of 215 pounds per acre for surface soil alone, but a loss of only 48 pounds where the absorption effected by the subsoil is introduced as a factor during the second year of the experiment. It is thus apparent that the presence of the underlying subsoil either inhibits the formation of magnesium sulfate within the surface soil, or, if not inhibiting its formation, increases the holding capacity of the soil for sulfur, as well as for magnesium; or else the subsoil absorbs not only the basic ion but also the acid radicle. The first assumption is not tenable, and it is probably beyond doubt that the subsoil has effected an absorption of both Mg and SO₄ ions without any interchange of bases, this being confirmed by the analyses of the alkalies in the leachings.

Considering the treatments in the order in which they occur in table 1, the 8-ton applications of burnt lime slightly depress the amounts of sulfur coming through in the leachings, as compared with the other and equivalent treatments; but *the 32-ton and 100-ton treatments practically inhibit the outward movement of sulfur in solution*. No such retardation in the sulfate leachings was demonstrated by the precipitated carbonate or by the natural carbonate of lime, even in the case of the 100-ton-equivalent applications. It may be observed that during the second year, when the 32-ton treatment of lime had become in large part carbonated, the increase in the sulfates leached was over six-fold. It is hoped that we may be able later to state more definitely whether the marked depression is due to chemical or to biological causes, or to a combination of the two agencies.

The effect of oxide of magnesium has been the reverse of that produced by burnt lime. Either the tendency to form sulfates was engendered or accelerated by magnesia, or else the ability of the soil to retain sulfates has been depressed. As previously stated, we have observed that the movement of sulfur and that of magnesium are parallel in the leachings so far studied and it is interesting to note that in the case of the 32-ton treatments of both oxide and carbonate of magnesium, the magnesium appeared in the leachings from the subsoil lysimeters in appreciable amounts prior to like appearance in the case of the 100-ton treatments. The amounts of sulfur leached from these same tanks were in accord with this unanticipated finding of magnesium.

All of the natural carbonates, in the several amounts, appeared to bring about conditions which caused an augmented outgo of SO₄, when compared with subsoil tanks which received no carbonate treatment. The concen-

trations of the leachings during the second year were greater, with reference to their sulfate contents, than during the first year, but the volumes leached during the second year were less than during the initial annual period, so that, as a result, when calculated to the pounds-per-acre basis, the actual amounts of sulfur leached during the second year were smaller than those which passed through during the first year. If the abnormal losses are due to biological activation, such a result would be expected.

The differences between the results produced by the natural and the precipitated carbonates are interesting. The lesser solubilities of the natural carbonates are reflected both in the lack of appreciable increase in the amounts of SO_3 leached and in the constancy of the碱inities of the several leachings. The greater active mass afforded the soil for the satisfying of its absorptive power, in the case of the more soluble oxides and precipitated carbonates, has caused an earlier demonstration of the accelerated movement of alkali-earth sulfates, if, indeed, the natural carbonates are ever to demonstrate such a tendency.

The soil's initial total sulfur trioxide content was 0.1290 per cent, while the subsoil showed a total per cent of 0.1249. The amount of SO_3 extracted by a 3-hour agitation with distilled water was 0.0152 per cent for the surface soil and 0.0036 per cent for the subsoil. Twenty-gram charges were used in making both determinations.

It can readily be seen that a continued loss of sulfates, in proportions approximating those which have transpired to date, would effect a speedy and absolute depletion of the initial organic sulfur content. Particularly is this true of the oxide and precipitated carbonate of magnesia, while, to date, the excessive burnt-lime treatments are an outstanding exception. In addition, however, to the initial store of organic and inorganic sulfur, there must needs be considered the excessive supplementary aerial supply.

The University farm is located within the boundary of the city of Knoxville, where a large amount of soft coal is burned. This may be appreciated by the fact that the average of analyses of three years' rainfall by three analysts shows the annual precipitation of sulfur trioxide to be 124 pounds per acre. The analytical data obtained from analyses of the original soil, long exposed to the sulfur of rainfall, do not suggest any characteristic property of this soil to conserve the sulfur obtained from aerial sources.

PLATE 1

FIG. 1. OUTSIDE VIEW OF HILLSIDE LYSIMETER EQUIPMENT
FIG. 2. INSIDE VIEW OF HILLSIDE LYSIMETER EQUIPMENT

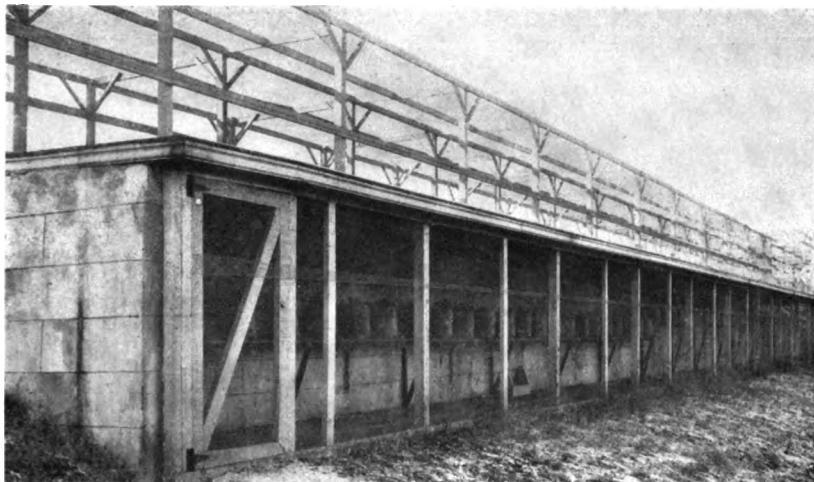


FIG. 1

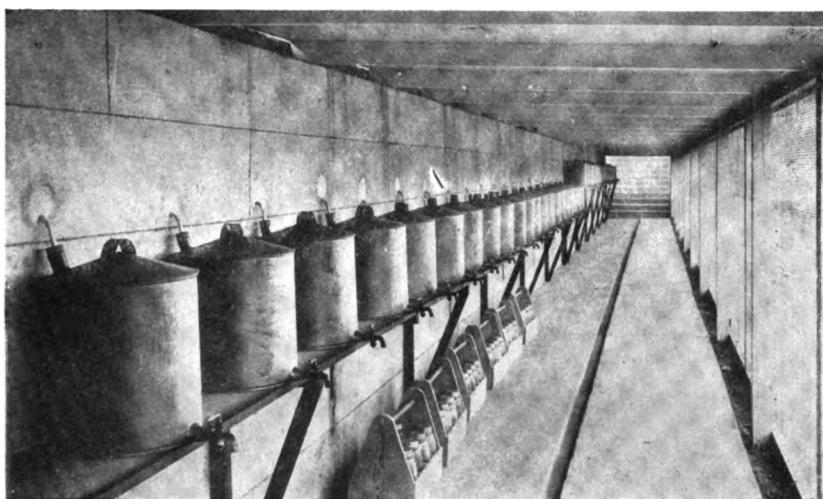


FIG. 2

THE RATE OF WATER MOVEMENT IN AERATED SOILS

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Of the many processes operative in soils that are of interest to plant physiologists, those concerned in the movement of water are of primary importance, because they modify the effects of all the other processes upon rooted plants. Furthermore, as has been repeatedly emphasized (5, 14, 15, 16, 17) the ratio between the rate at which water moves from the soil into the plant and the rate at which it moves from the plant into the atmosphere is often of supreme importance in conditioning the existence of a given plant or species of plant in a given habitat. Before any quantitative information of general applicability can be obtained regarding the rates at which plants absorb water from the soil, the preliminary question dealing with the maintenance of intimate contact of absorptive surface and soil water must be answered. Of the two ways in which this contact may be effected, namely, by growth of the roots themselves through the soil as the water is absorbed, and by movement of the soil moisture to the roots, only the second process is susceptible of investigation independently of the other. It thus appears that quantitative knowledge concerning the movement of soil water is of immediate importance to the general physiology of rooted plants. Since *quantitative* knowledge concerning the rates of movement of soil moisture is desired it is important that a definite, quantitative hypothesis concerning water movement be developed, and this is attempted in this paper. The results, combined with experimental data, will, it is hoped, be of service in the elaboration of a more precise, mathematical statement from which the movement of soil moisture may be calculated from measurable soil characters.

In approaching this problem of the movement of water in soils it appears advisable to narrow the field of initial inquiry to non-toxic, non-saline, drained (and aerated) soils because the greater number of higher plants of temperate zones (those that are at present economically and ecologically the most important) are rooted in such soils. The processes by which water movement takes place may be grouped under two heads: mass, or molar movement of water, and molecular movement. The latter, which includes movement by local evaporation and condensation and by diffusion through the soil solution, has, in general, a relatively small effect upon any given plant during its growing season. In the following discussion only molar movement of water will be considered.

The solution of this problem has been greatly furthered by the work of Briggs (6, 7) and of Buckingham (10). Although these authors obtained no satisfactory quantitative statement of the movement of water, chiefly because they possessed no instrument suitable for measuring rates,¹ their descriptions of the structure of drained, water-containing soils and their very complete analysis of the forces operative in mass movement of water form a valuable basis for such a statement. Obviously, the immediate need is for an instrument that will be suitable for absorbing water from field soils, an instrument whose maximum rate of absorption is greater than the rate of water movement in the soil, whose contact with the soil is intimate and whose absorptive surface remains fixed in position. The high maximum absorption is essential in order that the water may be absorbed as fast as it reaches the instrument so that the rate of movement of soil moisture may be determined. The fixed position of the absorptive surface is essential in order that movement of this surface shall not complicate the analysis of the results of its use, or, as when air is used as an absorbing medium, a dry layer of soil shall not be formed, which diminishes the rate of absorption.

The diffusion of water into a solution from which it is separated by a septum that permits the counter-diffusion of the other components of the solution to only a slight degree has been shown by Pulling and Livingston (19) to be of potential value as the operative principle of such an instrument and was employed in this investigation. The collodion membrane, prepared in the manner described by Bigelow and Gemberling (4) and by Smith (21) was found to be adequate, although a 12-hour immersion in an 80 per cent solution of ethyl alcohol in water, after a certain amount of drying, was found to decrease the permeability and increase the uniformity of the membranes.

In the present studies magnesium sulfate solutions were substituted for those of cane sugar, which were used in the earlier work, because of the high viscosity, great variation in viscosity with temperature changes and chemical unstableness of the latter. Preliminary experimentation, the details of which are not germane to the present discussion, led to the selection of magnesium sulfate in solutions of approximately M.1.5 concentration.² As high a concentration as this is not necessary to remove water from the soil, but in order that the instrument might operate for several hours with uniform, water-absorbing power, it appeared necessary that the difference between the specific gravities of the portion of the solution diluted by entering soil water and that of the undiluted portion be great enough to insure rapid operation of convection currents, thus rendering a higher concentration necessary.

The glass thistle tubes used in the Tucson experiments (19), although they

¹ The difficulties of obtaining proper contact between absorbing surface and soil seem to have prevented field tests with the instruments that had previously been described. See Briggs and McCall (8), Livingston (14) and Whitney and Cameron (22).

² This solution was obtained by dissolving 1.5 of the gram-molecular weight of hydrated (approximately 7 H₂O) magnesium sulfate in 1000 cc. of water at approximately 20°C.

were useful in the preliminary tests, were unsuitable for field use because of their fragility and clumsiness. The form of the instrument that was used

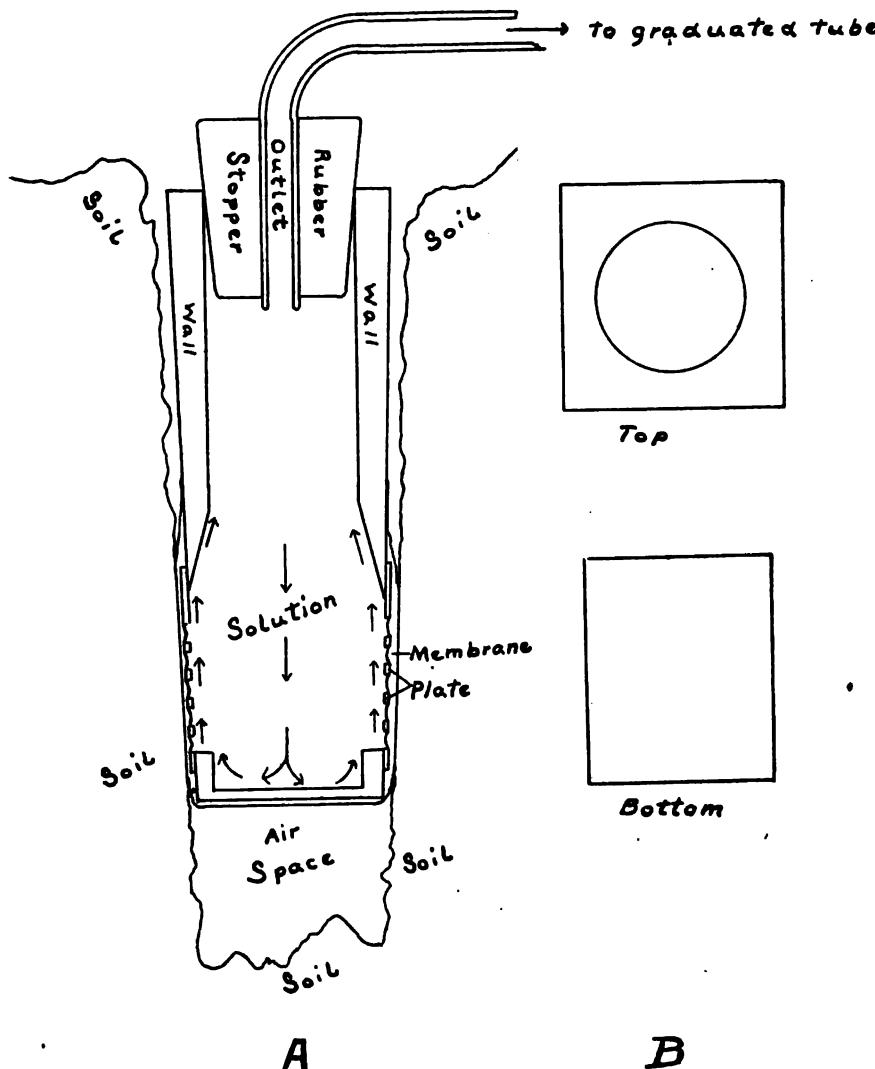


FIG. 1. THE OSMOMETER

A, Sectional view of the osmometer in position in the soil. The arrows show the approximate general direction of convection currents. B, Top and bottom views of the osmometer.

in the present field tests is shown in figure 1. Each instrument was made from a bar of brass, square in cross section. This bar was planed on two opposite faces to form a wedge, $1\frac{1}{2}$ by $1\frac{1}{2}$ inches at the top and $1\frac{1}{2}$ by 1 inch at the

bottom. A cylindrical core, $\frac{7}{8}$ inch in diameter, was removed longitudinally from the larger end almost to the smaller, and a circular opening, also $\frac{7}{8}$ inch in diameter, was then bored through each of the planed faces, opposite each other and near the lower end, thus communicating with the central cavity. Experimental search for a means to control convection currents (in lieu of stirring) sufficiently to obtain fairly constant rates of intake when the instrument was immersed in water led to the selection of the following as the most convenient. Thin, brass plates, perforated as extensively as possible with holes $\frac{1}{8}$ inch in diameter, were soldered over the lateral openings. Although a single membrane was applied over each entire plate, communication with the soil was virtually effected through many small membranes.

To place the osmometers in position in the soil a trench, narrower than the instrument and deeper than its height, was cut with a sharp knife and the soil removed. The trench was widened until it was only slightly narrower than, and of the same sectional shape as the instrument, which was then carefully lowered into it and gradually, during the course of fifteen or twenty minutes, pushed deeper until further movement was not followed by an increase in rate of water intake, when it was assumed that the greatest possible area of membrane was in contact with the soil.

An objectionable feature of this form of instrument is the flat surface to which the membranes are applied, for the probability that a membrane will adhere perfectly to every part of it is small. A slight trace of moisture remaining between the membrane and the brass surface forms a drop into which salt gradually diffuses as the instrument is used. Into this solution water diffuses, enlarging the drop and slowly separating brass and collodion until connection is established with either the interior or the exterior. If it be with the former, the process proceeds until a continuous passage is formed, underneath the membrane, between the interior and the exterior of the instrument. Such leaks may be so small as wholly to escape notice while the rate of water intake is great, as in trials of the instrument against water, but may produce large errors when the rates are small, as in many cases of its operation in the soil.

Of the many methods tried for preventing errors caused by these leaks the following was adopted. Instrument and delivery tube were filled with distilled water and the instrument placed in a small vessel containing water, the upper level of which was above that of the membrane; the graduated delivery tube was elevated until the water meniscus in the tube stood 13 cm. (uniformly maintained but selected from considerations of convenience only) above the water level outside. The rate at which water passed outward through the membrane (or other accidental exits) as indicated by the fall of the column in the tube, was considered an index of the instrument's permeability. After considerable experimentation it was decided to discard all membranes that permitted a loss of water at a rate greater than one cubic millimeter per 5-minute interval. This limit is perhaps so low that many

serviceable membranes were discarded, but as it appeared to be a certain insurance against leaks it was believed best to adopt it. Instruments were tested frequently and whenever one was found that exhibited greater permeability than at a previous test it was considered unsatisfactory and all the data acquired with it during the interim were discarded. Because of these suspected leaks data from less than a third of the original number of tests can be presented.

Turning to a consideration of the movement of water in drained soils it will be of advantage to picture the process descriptively. In such soils

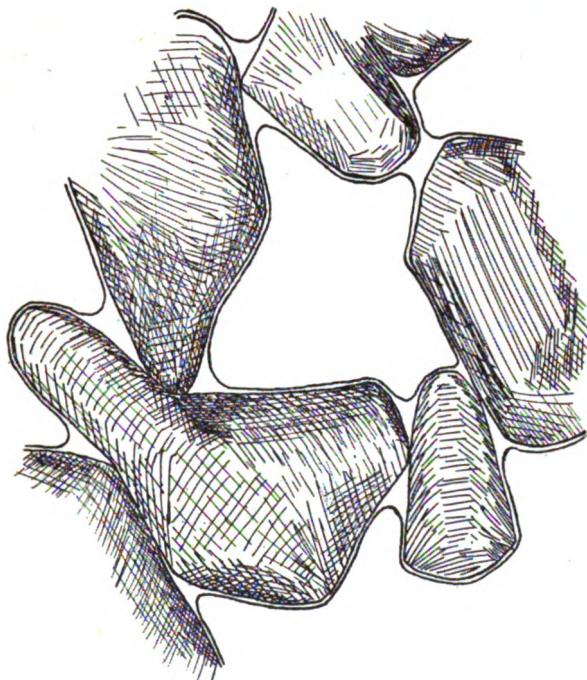


FIG. 2. HYPOTHETICAL SECTION THROUGH A DRAINED SOIL, SHOWING WEDGES AND FILMS

water exists (fig. 2) in the form of thin films covering the soil grains and confluent with masses of water that are of great diversity of form although alike in having an air-water interface concave to the air. Because of their general sectional shape these masses have been termed "wedges" by Buckingham (10) and this designation will be used in the following to distinguish them conveniently from the hygroscopic films that cover the soil grains. Water of hydration, the so-called colloid water and water that has penetrated the minute cracks in the soil grains need not be considered since the water-holding forces that are operative in these cases are so powerful that for present purposes the water may be regarded as virtually a part of the soil grain with which it is

associated. It is necessary only to consider water in the 'forms.'³ mentioned in the text books as capillary and hygroscopic water, for the third of these forms, gravitational water, is assumed absent since the soil is drained.

As far as plants are concerned the hygroscopic water is of little importance, for, as has been repeatedly shown, they are unable to withdraw it from the soil, as, indeed, a consideration of the elementary principles of equilibrium will demonstrate, and it cannot be reduced in amount by molar water movement. The usual text-book statement is that the "water films" increase and decrease in thickness with varying amounts of soil moisture. The results of investigations on the flow of water in capillary tubes has shown, however, that slipping in the layer of liquid adjoining the wall does not take place, which indicates that relatively strong forces hold it in position. Also Bakker's (1, 2, 3) researches indicate that the thickness of the capillary film is slight (about 1.6×10^{-7} cm.)⁴ and practically invariable, small variations resulting only in response to large changes in vapor pressure or temperature. Since it is so thin, this film is, throughout its thickness, a fusion of both the gas-liquid interfacial layer of the wedges (the surface tension film) and that water layer of the wedges that immediately adjoins the soil grain. Thus the surface forces resident in the gas-liquid interface of the wedges are transmitted by this hygroscopic film to the soil grains so that the soil mass acquires structure.

In concave surfaces the capillary tension⁵ varies inversely as the radius of curvature, becoming least when this is infinite, i.e. when the surface is plane. Since capillary tension is the force retaining water in the wedges, it is evident that for all parts of the soil to be in equilibrium with regard to water movement, the radii of curvature of the concave surfaces (as Briggs long since pointed out) must be equal throughout, regardless of the actual amount of water in any individual wedge or in any number of them. The

³ As Free (12) has pointed out, references in this connection to forms of water are very objectionable because of the implication that a change of condition or state accompanies change from one form to another, whereas, so far as is known, the difference is solely in the forces (quantitative or qualitative) holding the water in a given position with reference to the soil grains.

⁴ This value for the thickness of a capillary film is smaller than that usually given, which ranges from $5-10 \times 10^{-7}$ cm. Quincke (20) gave the distance through which a glass wall attracts water as about 5×10^{-6} cm., a much larger value than that of later investigators. The actual thickness is of little importance in this connection since the order of magnitude is practically the same for the different values. For further references and a brief statement of the range of attraction of molecules, see Lewis (13).

⁵ *Surface tension* may be defined as the tension per centimeter in the plane surface of a liquid under a given set of temperature and pressure conditions. *Surface energy* is defined as the product of surface tension and surface area. *Capillary tension* is used in this paper to designate the tension, often confused with both the foregoing, brought about by the curvature of a surface that possesses surface tension. Surface tension is thus considered as a property of liquids, varying only with their nature, with temperature and with imposed pressure, while capillary tension varies not only with the surface tension but also with the curvature of the surface.

withdrawal of water from any one wedge will result, in general, in a decrease in the radius of its surface curvature, since almost invariably the space occupied by the water is narrower near the point of contact of the grains than near the air-water interface. This increase in curvature will be accompanied by an increased capillary tension as mentioned above.

A general description of the movement of water in drained soils may now be attempted. As the capillary tension increases the effect will theoretically be two-fold: first, adjoining soil grains that are not in contact (fig. 2) but whose hygroscopic films are a continuation of the surface-tension layer of the wedge and transmit tensions to the grains, will be drawn closer together; and second, water will be moved from adjoining wedges with less curvature (i.e., with greater radius of curvature). In field soils the former process will, in general, be negligible at and below the depths occupied by even shallow-rooted plants so long as forces of greater magnitude than those of capillary tension (such as the expansive force of freezing water) are not active. In the early spring in regions where the soil freezes, in newly-ploughed ground, or on the surface of a dried soil recently wetted, rearrangements of the soil grains would of course be of significance. In general, however, in the deeper portions of the soil the response to the increased tension is movement of water only. Suppose a wedge to be in direct communication with others, the invariable condition in moist field soils of the type considered in this discussion, and suppose water to be removed from the first wedge. Because of the increased capillary tension in the surface of this wedge, water will move into it from the others, through the communications. If these were the only wedges concerned, the first would not obtain sufficient water to compensate for the loss, because movement could proceed only until the curvatures of all surfaces involved became equal, each wedge consequently containing less water than before. If a third set be now considered to be in communication with the second, it is evident that water will move from these wedges to those of the second set, whose radius of curvature becomes thus greater, enabling the first wedge to obtain more water. As additional sets are added to this chain the degree to which the first wedge may, in time, approximate its original water content increases, so that in a field soil, if the amount of water withdrawn is relatively small and the time for recovery is relatively great, the wedges initially depleted will acquire an amount of water practically equal to that originally lost.

If the water is removed, however, by an absorbing body that acts with a continuous and uniform force of absorption, which is, however, not of sufficient intensity to break communication between the wedges, the condition of non-equilibrium should be constant in that portion of the soil immediately adjoining the absorbing body. At any instant of time the air-water interfaces of the wedges touching this body will have the least radius of curvature of any in the soil mass, supposing the soil mass to have been originally in equilibrium. The interfaces of the next wedges will have a greater radius of curvature and the radius will increase from one set of wedges to the next until the unaltered

soil is reached. As time progresses, the distance through the soil in which the curvature of the surface has been altered, will increase. Since the curvatures of the surfaces in contact with the absorbing body remain constant (because the absorbing body acts with a uniform and continuous absorptive force), the wedge and the absorbing body remaining in practical equilibrium, it follows that the difference between the curvatures of the air-water interfaces of any one wedge and the next will continually decrease, since the number of wedges whose curvatures are unequal will increase with time between the fixed limits of curvature set by the conditions at the absorbing body and the unaltered soil. Since throughout the affected portion of the soil the curvatures become more and more nearly equal as time progresses, and since the movement of water is conditioned by the difference between the surface curvatures of adjoining wedges, it follows that the rate of water movement should become less and less. It is also to be expected that the decrease in rate will be most evident when the number of wedges affected is small. If, then, the rates of water movement in the soil were plotted against the corresponding points in time as abscissas, the resulting curve should exhibit a steep slope during the first few intervals, the rates decreasing more and more gradually as time proceeds, until the curve becomes almost parallel to the horizontal axis, upon which the intervals of time are plotted. All graphs obtained with the osmometers exhibit this latter characteristic, those extending over the longest periods of time showing most plainly the increasing constancy in the rate of movement.

It is patent, however, from an inspection of the graphs that, although they are similar in their more general aspects, there are many particulars in which they differ. Some are higher than others, some begin with larger rates than those appearing later in the graph, others with smaller rates; in some the decline is very rapid, in others it is much more gradual. The interpretation of these experimental graphs is divided into four sections. The first deals with their general slope, i.e., the rates at which movement of water diminishes with time. The second deals with the heights of the graphs, i.e., the actual amounts of water moved through equal, fixed planes per unit of time. The third deals with the observations made during the first few time periods of the tests. These initial rates appear to be controlled in their magnitude by conditions other than those considered in sections 1 and 2. The fourth deals with the apparent fluctuations in the rate of water movement.

1. The Slopes of the Graphs

The movement of water through the soil is opposed by frictional and capillary resistances. Since these resistances are chiefly due to the existence of the soil water in discrete masses it follows that the greater the number of such masses the greater should be the total resistance, and the slower should be the rate. It also follows that if the number of these wedges is initially

great in proportion to the amount of water held, the addition of a unit number will produce less effect than if the number is initially small. As the chief resistance to water movement is probably encountered in moving from one wedge to another, it would appear that the greater share of the time should be consumed in movement between wedges, and not in them. Movement of water must accordingly be pictured as proceeding discontinuously from wedge to wedge, so that a steady state of water movement can never be set up in the soil, although as the rate approaches zero the effect of the lag will be less and less noticeable. So it appears that a consideration of the numbers and volumes of the wedges per unit of gross soil volume is necessary before the preceding general statements can become useful.

Since the wedges are formed about points of contact of soil grains, it is at once obvious that their number will, in general, depend upon the size, shape and number of soil grains in a unit of gross or field volume, since the number of contacts is determined by these variables. In any given case the addition of soil grains in the interstices of a soil mass will result in an increase in the number of wedges, the actual increase depending not only upon the number, size and shape of the added soil grains but also upon the number, size and shape of those already present. Although it appears unprofitable to attempt the formulation of a relation connecting these variables, it seems possible that experimentation may furnish an integration of their various effects as a single, measurable magnitude.

Such experimentation has been made and, although the details must be reserved for another article, the general reasoning and some of the results are of importance in this connection. In a unit volume of any given sample of soil the number of soil grains may be considered to be directly proportional to the weight of the dry soil present in that volume. This would probably be an unjustified statement if very small volumes of a soil consisting of unequal grains were considered. But in a sample having a volume of several cubic centimeters these differences are negligible for ordinary, arable soils, so that these soils may be considered as made up of *average grains*, equal weights of dry soil signifying equal numbers of grains. If two equal volumes of such a soil have the same number of soil particles and the same water content, the number of wedges should be the same, the curvatures of the air-water interfaces should be the same, and water should not move from one sample to the other if they were placed in contact. For soils of heterogeneous composition this statement will be the more nearly true the smaller the amount of water the samples contain; for with small amounts, the water is more closely disposed about the points of contact of the grains.

If samples of some one soil are graded according to their solid content and according to their liquid content, each sample, however, having such a water content that the curvatures of its water surfaces are the same as those of the others (thus rendering their capillary tensions equal), the two series of values (solid content per unit of volume and corresponding liquid content) obtained

may be plotted one against the other to form a graph that may be regarded as exhibiting the variation in the number of wedges per unit of gross soil volume, corresponding to changes in the number of soil grains per unit of volume. If the curvatures are all the same and the water contents are the same no sample should be able to obtain water from any of the others and each should deliver water at the same rate as the others to a standard absorbing body.

A method has been devised by which such data may be obtained. By its means samples of a given soil, each of approximately uniform packing may be secured that permit the movement of only about 0.001 gm. of water during 24 hours through an area of about 30 sq. mm. to a standard absorbing body. The liquid and solid contents are then determined for each sample as weights per unit of gross volume. If the weights of solids per unit of gross volume are plotted for the several samples as abscissas, the corresponding weights of liquid plotted as ordinates, and the points so obtained connected by a line, a curve is produced that is characteristic for the particular soil investigated. Each soil is thus considered as a three-phase system; solid-liquid-gas, that is defined in any condition of packing and water content when two of the three variables are stated in reference to the entire volume occupied by the three. These variables should, of course, be stated in terms of volume but for the present the more convenient weights will be used.

By changing the absorbing power of the standard body a family of such curves might be obtained. These curves will be called the liquid-solid curves, or, more briefly, the l-s curves characteristic of the particular soil from which the data were derived. Each curve may be considered as exhibiting the change in the number of wedges per unit of gross soil volume that occurs as the number of soil grains is altered, the curvatures of the air-water interfaces remaining the same. The lowest curve, which is the only one that has been determined, also exhibits the water content for each solid content of the given soil below which the rate of water movement can be considered negligible. Because of its importance this curve should be specially designated, and since it is the lowest of the family it will be termed the α l-s curve, or, briefly, the α curve, and the water content corresponding to any ordinate value thereon will be termed the α water content of the soil for the corresponding solid content per unit of gross volume.

A statement of the rate of decrease in the amount of water moving through a given area in the soil in unit time into an absorbing body that removes the water as fast as it reaches the body (as an osmometer or root hair may be supposed to act) may now be attempted. On the basis of the preceding reasoning this rate of decrease will vary inversely with time and also inversely with the α water content for the particular solid content of the soil that was encountered in the test considered. Or

$$\frac{dy}{dt} = k/m t \quad (1)$$

In which k represents a factor of proportionality, m , the proper α water con-

tent, and y , the decrease in the rate of water movement during the time, t , reckoned from the beginning of the test. For any given test m is a constant and since k is also a constant the equation may be integrated directly:

$$y = k/m \cdot \ln t + C \quad (2)$$

In which C represents the constant of integration, \ln is the accepted abbreviation for "natural logarithm of" and the other symbols have their former significance.

The rate of *water movement* at any instant of time may then be expressed by the equation

$$dx/dt = s - (k/m) \ln t + C \quad (3)$$

In which x represents the amount of water that has moved through the given plane from the beginning of the test until the time, t ; s , the amount that moved through the plane during the first interval of time, an amount that *should* move across in each interval if no decrease in rate occurs. The other symbols have their former significance.

To render equation (3) suitable for experimental verification it must be integrated for a finite period of time. Since, for any one test, s is a constant the integration may be readily performed:

$$x = st - (k/m)t(\ln t - 1) + Ct + C'$$

In which C' is the new constant of integration and the other symbols have their former significance.

When $t = 0$, $x = 0$ and accordingly $C' = 0$

When $t = 1$, $x = s$ and since s is defined as the rate during the first interval of time

$$x = s + (k/m) + C$$

for during the first interval of time $t = 1$ and $\ln 1 = 0$. Accordingly

$C = -(k/m)$, and the complete equation becomes

$$x = st - (tk/m)(\ln t - 1) - tk/m$$

or, rearranging,

$$x = st - (k/m)t \cdot \ln t \quad [\text{see footnote 6}] \quad (4)$$

For any unit interval of time,

$$x_2 - x_1 = s(t_2 - t_1) - (k/m)(t_2 \ln t_2 - t_1 \ln t_1)$$

* It will be at once noted that the effects of temperature, changes in viscosity and surface tension, etc., are not considered in the equation. While these variables undoubtedly influence the rate of water movement, it seems inadvisable to consider them in a preliminary study. The data secured in these tests indicate, moreover, that these variables have slight effects compared with fluctuations in the rates and experimental errors, so that it is necessary to ignore them.

or since $t_2 - t_1 = 1$

$$x_2 - x_1 = s - (k/m) (t_2 \cdot \ln t_2 - t_1 \cdot \ln t_1) \quad (5)$$

or

$$s = x_2 - x_1 + (k/m) (t_2 \cdot \ln t_2 - t_1 \cdot \ln t_1) \quad (6)$$

To test the equation, k must be determined and this can be done only with approximate success from the data at hand because of the lack of knowledge concerning the initial rate, s , and because of the fluctuation from one period to another in the amount of water absorbed by the osmometer. The physical meaning of these fluctuations will be discussed later, and for the present purpose a smoothed curve will be considered. This smoothed curve should represent the same total movement of water during the entire course of the test as that found by experiment but it should have no inversions, i.e., no ordinate value should be higher than any preceding ordinate value. The fluctuations are thus temporarily looked upon as accidental or non-essential deviations of experimental observations from the true course of a physical process.

Since k has the same value for all tests, at least for each soil, the one exhibiting smaller fluctuations in the observed rates than do the others, should be selected for its determination. Two values for the rates are needed and these are best selected toward the end of the test when the decrease in rate is small, in order that the smoothing may be done with the least error. Constants obtained in this way are, of course, not accurate but the importance of the subject and the difficulty of making field tests justifies the use of what data may be secured even though, from the standpoint of strict accuracy, they may appear inadequate.

Test No. 3 was selected and the rates were plotted between period 30 and the end of the test, period 54. A straight line was then drawn as nearly as possible along the course it was considered the smoothed curve should pass. At period 35 this line had a height of 5.5 units, and at period 52 a height of 5 units. Since x represents the total amount of water absorbed by the osmometer since the beginning of the test to the time at which x is measured, $x_{t=35} - x_{t=34} = 5.5$ cu. mm. and $x_{t=51} - x_{t=51} = 5$ cu. mm. Data for two simultaneous equations of the form of equation (6) are thereby presented for the determination of k :

$$\begin{aligned} s &= 5.5 + (k/m) (t_{35} \cdot \ln t_{35} - t_{34} \cdot \ln t_{34}) \\ s &= 5.0 + (k/m) (t_{52} \cdot \ln t_{52} - t_{51} \cdot \ln t_{51}) \end{aligned}$$

Combining these equations and rearranging terms

$$5.5 - 5.0 = (k/m) (t_{52} \cdot \ln t_{52} - t_{51} \cdot \ln t_{51} - t_{35} \cdot \ln t_{35} + t_{34} \cdot \ln t_{34})$$

or

$$\frac{m (5.5 - 5.0)}{t_{52} \cdot \ln t_{52} - t_{51} \cdot \ln t_{51} - t_{35} \cdot \ln t_{35} + t_{34} \cdot \ln t_{34}} = k$$

By the method of experimentation already discussed to some extent m , the α water content of the soil, was found to be 76 cubic millimeters for each cubic centimeter of this soil when the solid content per unit of gross volume was the same as that found from volume samples of the field soil taken when the osmometer test was made. Substituting this value of m and the proper values for the other symbols

$$\frac{76 \times 0.5}{205.4676 - 200.5218 - 124.4390 + 119.8976} = k = 94$$

In the graphs (fig. 3-11) the continuous lines represent the results of calculating $x_2 - x_1$ for the various time periods and the dotted points represent

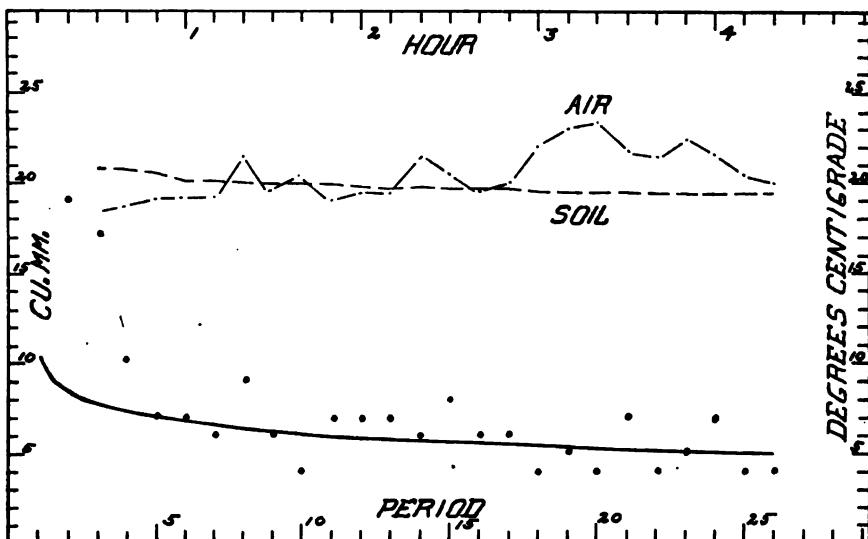


FIG. 3. TEST 1 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL
Total percentage of moisture on the basis of field volume, 0.193 per cent.

tubes of the osmometers. Remembering that this section deals only with the general *slopes* of the curves, the discussion of their *heights* being reserved for section 2, it will be noted that the graph of the fourth test (fig. 6) is the only one that has a slope notably different from the actual osmometer readings. In this test 0.08 cm. of rain fell during the hour and a half between period 3 and period 12. During the rain the osmometer was covered with canvas which also prevented direct wetting of the soil within two feet of its position. This is of significance when the rates of water intake are observed between periods 13 and 32. About thirty minutes after the cessation of the rain the rates decreased greatly and continued to do so more gradually until period 32 when they became more nearly constant. The speed with which the saturated surface soil attained equilibrium with the dryer soil surrounding

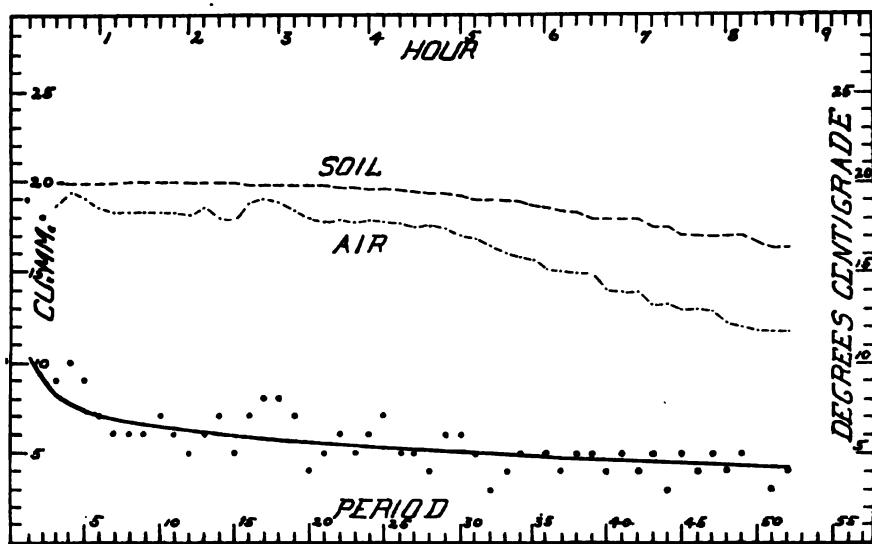


FIG. 4. TEST 2 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL
Total percentage of moisture on the basis of field volume, 0.200 per cent.

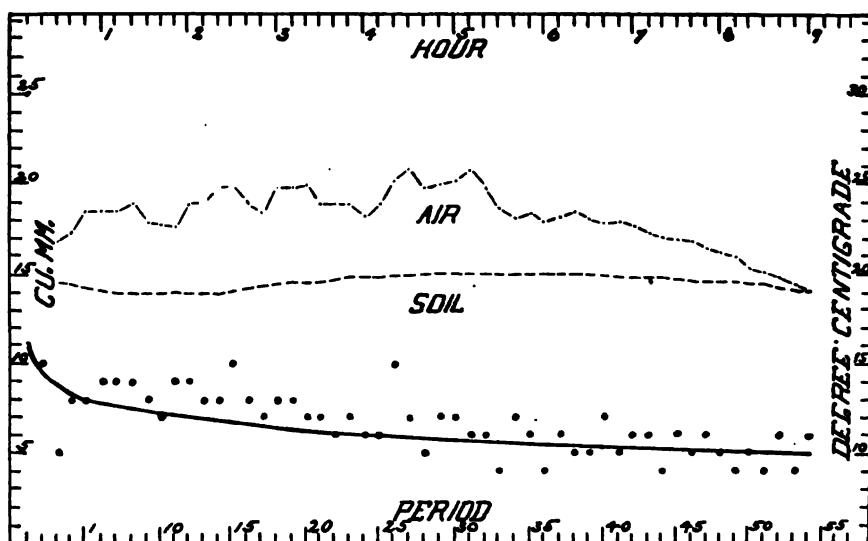


FIG. 5. TEST 3 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL
Total percentage of moisture on the basis of field volume, 0.213 per cent.

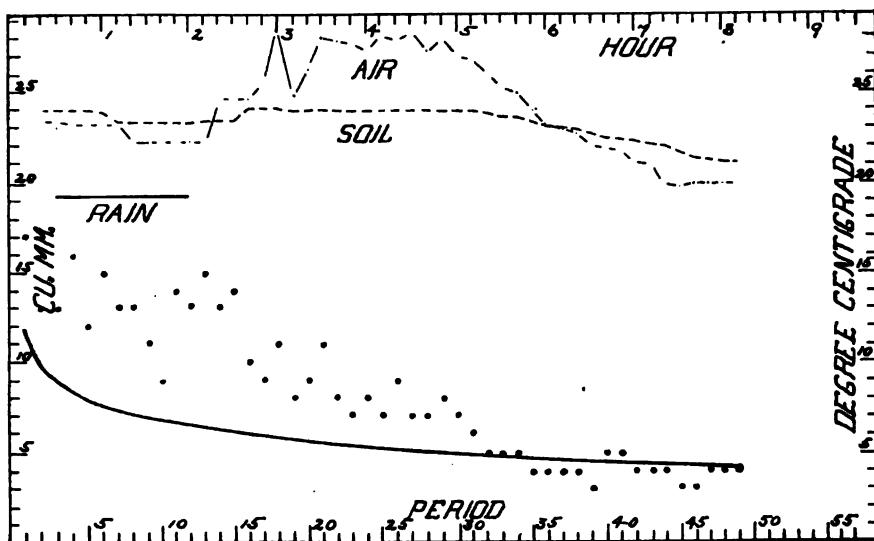


FIG. 6. TEST 4 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL
Total percentage of moisture on the basis of field volume, 0.211 per cent.

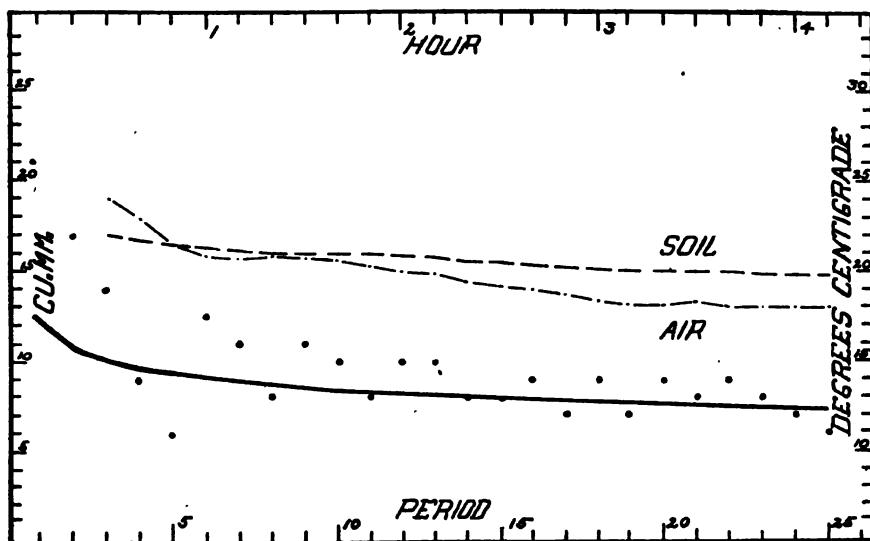


FIG. 7. TEST 5 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL
Total percentage of moisture on the basis of field volume, 0.237 per cent.

the corresponding water intake in cubic millimeters as read on the graduated osmometer (about 2 feet away and about 4 inches below the surface) is indicated by the general decrease in water intake between periods 13 and 32. Unfortunately the rain began so soon after the osmometer was placed in position that conclusions regarding the apparent increase in water intake are not justified, for, as will be explained in section 3, the first few observations on the rates in each test are unreliable.

Although in all cases the general decrease in the rates of water intake as time proceeds are very closely paralleled (with the exception noted above) by the slope of the calculated curve, it is conceivable that the conditions controlling the decrease in rates are wholly different from those considered in formulating the expression by which the calculated curve was obtained. That results calculated with the aid of an equation agree with experimental data is not conclusive evidence that the equation is based upon sound reasoning or justified assumptions. When, however, an equation is successfully applied to a given kind of process under conditions of great diversity, its value as a means of *predicting phenomena* must be recognized. Although the proposed equation has not by any means been thoroughly tested, it has been applied to a considerable range of moisture content for two soils of widely different character. The total volume percentage of soil moisture encountered in these tests ranges for the garden soil between 19.3 per cent and 26.0 per cent, and for the sand between 10.0 per cent and 21.5 per cent, while the value of m ranges between 51 and 77 for the garden soil and 51 and 61 for the sand (table 1). This expression, accordingly, may be considered to have considerable value for calculating the decrease in the rate of soil-water movement unless conditions during these tests were such as are not generally effective in the movement of soil moisture. Of such conditions three groups present themselves as possibly shaping the data: conditions introduced by the instrument, conditions of temperature and conditions of evaporation.

It is at first sight possible that during the operation of the osmometer the solution becomes more and more diluted by the entering water, each increment of dilution decreasing the osmometer's capacity to absorb water. During the preliminary experimentation in search of a method for increasing convection this question was very thoroughly considered. Table 2 presents the data obtained from a test of an osmometer constructed as described and figured in this article, operating against water for 11 hours and 20 minutes. The observations were made at intervals of 5 minutes (the intervals in the tests against soil were 10 minutes long) and is representative of the average magnitude and fluctuation of the readings for all the tests with this type of osmometer. They are presented because the time during which this test was made was greater than for any other. It is evident that there is no decrease in rate at all proportional to that observed in the soil tests even though the average intake during ten minutes was several times the rates for the soil tests. It must then be concluded that dilution of the osmometer

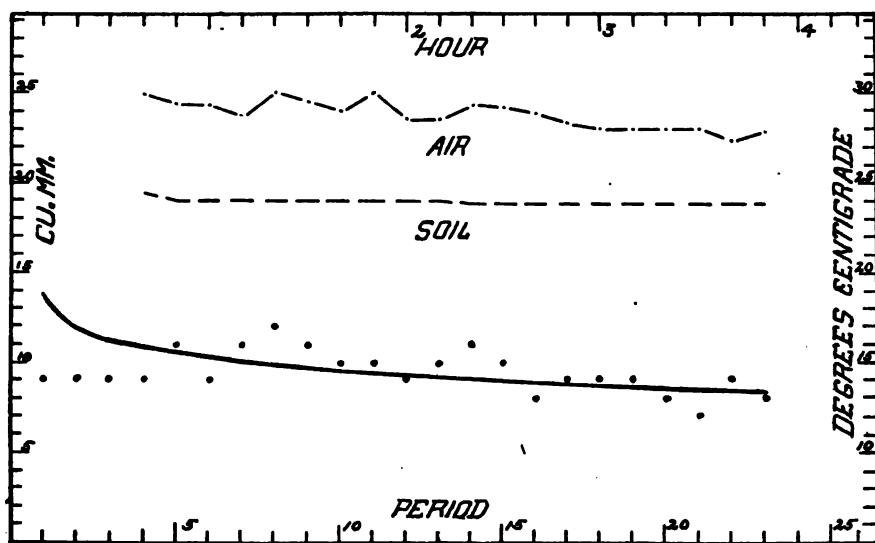


FIG. 8. TEST 6 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL

Total percentage of moisture on the basis of field volume, 0.260 per cent.

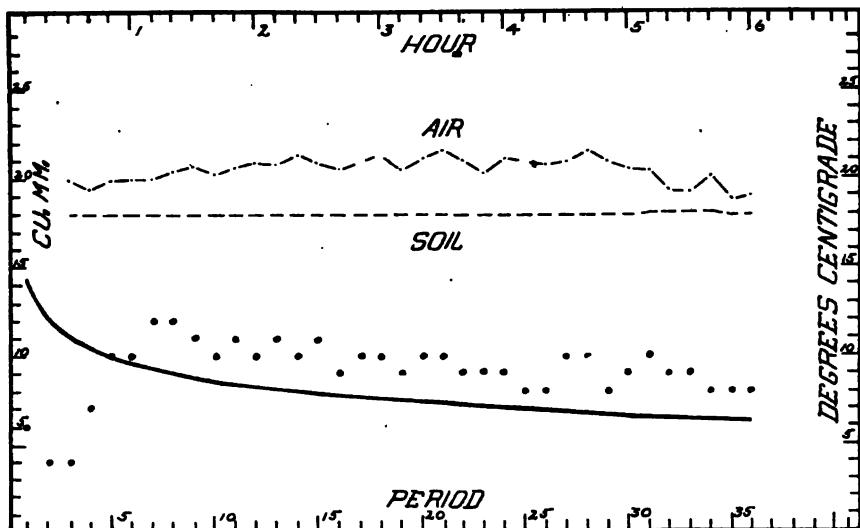


FIG. 9. TEST 7 OF THE OSMOMETER OPERATING AGAINST GARDEN SOIL

Total percentage of moisture on the basis of field volume, 0.257 per cent.

solution is not an explanation of the decrease in rate as observed in the tests with soil.

Since temperature is known to affect the equilibrium between solutions and pure solvent when the imposed pressure remains constant, it might seem that temperature changes could be the controlling influence in shaping the graphs (although this effect of temperature probably does not always proportionally affect the speed with which equilibrium is attained for in many cases this rate of solvent entrance may be controlled within wide limits by the membrane, a circumstance not usually considered when the relation between osmotic pressure and root absorption is discussed). There is, however, no evidence that this is true. The temperature curves are not followed by the graphs of osmometer rates, although sudden variations in temperature are often accompanied by corresponding variations in the rates. This latter effect of temperature will be considered in section 4.

The effect of evaporation may be discussed under two heads: the actual depletion of the soil by evaporation from its surface, thus lessening the amount of water available for movement, and the increase in a capillary tension that is transmitted throughout the soil and increases the resistance to water movement. There is little evidence obtainable from these experiments so far as the former is concerned, although there is much general ground for the belief that considerable water is withdrawn from the soil during the course of a day under many conditions. Although a dust mulch very effectively diminishes evaporation from soils, this condition probably was true only in tests 1, 5, 8 and 9, and in the latter two the withdrawal of water by the roots of actively transpiring plants may have offset the effect of the mulch. In tests 1 and 5 the decrease in rate is approximately that calculated, indicating no abeyance of the conditions occasioning the decrease, as would be the case if evaporation were the cause of the diminished rate and a soil mulch were present that prevented evaporation.

Test 4 indicates the extent to which added water influences the rate of movement and may throw light on the question of the effect of withdrawal of water. The day was cloudy and there was almost no wind. The fact that the observed rates near the close of the test are very close to the calculated rates and the speed with which the rates decrease after cessation of the rain, indicate that the water was rather quickly distributed and that the amount precipitated (0.08 cm.) was too small to alter the water content of the soil except locally. It is thus probable that a considerable part of the water that entered the osmometer between the first effect of the rain on the rate and period 30 or 32 was derived from the upper layers of the soil, which later attained approximate equilibrium with the deeper layers. The effect of the rain was thus probably only transitory and without any far-reaching effects. Attention should again be directed to the fact that no data exist for determining the rates *before* the rain and an error in the height of the calculated curve might render the above statements untrue.

The questions concerning a capillary tension transmissible through the soil as such, without concomitant movement of water, its increase by evaporation and the consequent augmented resistance to soil-water movement, may be discussed with the aid of more direct data. Evaporation on sunny days increases during the morning, reaching a maximum intensity early in the afternoon and decreasing toward evening. If the removal of water from the upper layers of the soil by evaporation produces an increase in the capillary tension which is transmitted throughout the soil mass without involving movement of water, it might be expected that movement of soil moisture would

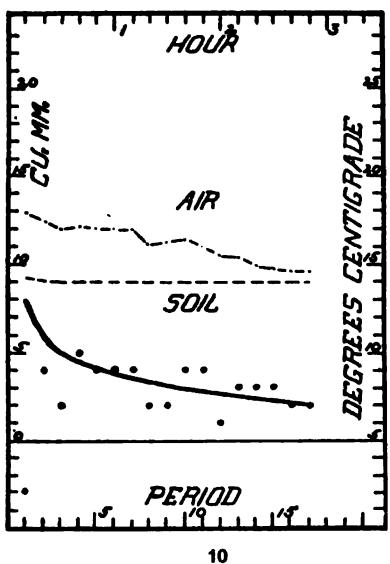


FIG. 10. TEST 8 OF THE OSMOMETER OPERATING AGAINST SAND
Total percentage of moisture on the basis of field volume, 0.100 per cent.

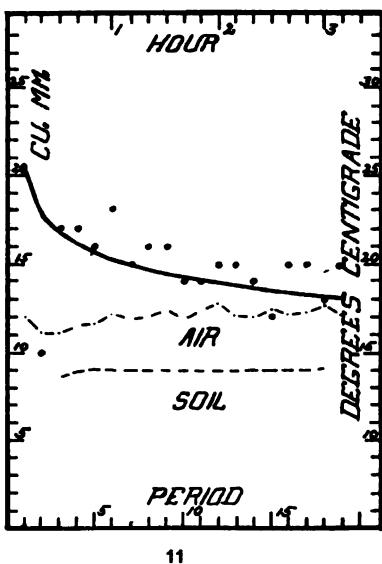


FIG. 11. TEST 9 OF THE OSMOMETER OPERATING AGAINST SAND
Total percentage of moisture on the basis of field volume, 0.215 per cent.

progressively decrease during the day. The rate of this decrease then, should be greatest during the early afternoon, or if it is assumed that the daily evaporation produces a mulch that decreases this noonday maximum, the diminution in rate should be more rapid in the morning. This latter suggestion finds support in the data from all the tests that were started in the morning, and these rates do not show a more rapid decline early in the afternoon as would be expected if the first of the two hypotheses were true. However, the rates in the tests that were begun in the afternoon exhibit the same sort of progressive decrease, and this does not seem susceptible of explanation on the ground of a far-reaching capillary tension. Since the decline in all cases is

very exactly accounted for in a quantitative manner by the general hypotheses of water movement, the capillary tension hypothesis advanced above may be dismissed, at least until more definite information of its effect is obtained.

2. THE HEIGHTS OF THE GRAPHS

An inspection of the graphs suggests at once that the amount of water in the soil is the chief condition controlling their heights. An analysis of the Tucson data (19) indicated that with a soil of a given moisture content, the rate of intake, as might perhaps be expected, varied with the area of the absorbing surface. This led to the idea that the initial rate of water movement depended upon the area of the membrane that was in actual contact with the water of the soil. The resistance of the soil to water movement would, according to this view, not affect the amount absorbed during the first instant of time but would subsequently diminish the rate. Part of the water in the soil must be considered to be held so strongly in position that it does not share in capillary movement. As already noted some of this water is held in the hygroscopic films (or "adsorbed" films) on the soil grains. In addition to this, water may be present in wedges whose air-water interfaces are so greatly curved and each wedge may be composed of so little water that a soil containing only this amount and that in the hygroscopic films can deliver water only at so small a rate that it may be considered negligible. Thus, only water that is present in the soil in excess of the sum of the two amounts just mentioned may be considered movable by capillary urge. This water content permitting but negligible water movement is termed the α water content of the soil and, as already mentioned, an experimental means for its approximation has been found.

If this view is correct, the initial rate of water absorption should be proportional to the $\frac{2}{3}$ power of the amount of water in unit field volume of a given soil in excess of the α content (i.e., proportional to the area of one face of a cube whose volume is that of the water contained in unit gross volume of soil in excess of the α water content). In mathematical form this statement becomes:

$$s = r(a - m)^{\frac{2}{3}}$$

In which r is a factor of proportionality, a signifies the amount of water present at the time of the test in a unit of gross volume of the soil and m represents, as before, the α water content of the same soil when it has the same content of solids as when a was determined. It is, of course, necessary that both a and m be determined by drying the soil to the same extent. Substituting this value of s in equations (4) and (5), the complete expressions, as exactly as it is now possible to formulate them, become

$$x = r(a - m)^{\frac{2}{3}} \cdot t - (k/m) \cdot t \ln t \quad (7)$$

$$x_2 - x_1 = r(a - m)^{\frac{2}{3}} - (k/m) (t_2 \ln t_2 - t_1 \ln t_1) \quad (8)$$

In order to evaluate r , s must be known for some one actual test. It was not possible to secure experimental values for s (section 3) but an approximation may be obtained by adding to the water intake during any interval of time the calculated amount that the rate had decreased since the beginning of the test, i.e., by substituting the proper values in equation (6). When this is done, using the data for time interval 52 of test 3 (the test that was used to evaluate k)

$$s = 5 + (94/76) (205.4676 - 200.5218) = 11.1 \text{ cubic millimeters, and}$$

$$r = s/(a - m)^3 = 11.1/26.6 = 0.417$$

The continuous lines in the figures for garden soil were plotted from equation (8) using this value for r and the value for k obtained in section 1. In general, the agreement is very close, the noteworthy exceptions being tests 4, 5 and 7. In test 4 the high rates previous to period 32 may probably be explained as an effect of the rain. For the others, unless there was an error in the determination of the moisture content of the soil, the expression is not an accurate statement of the rate of water movement under the conditions obtaining during those tests. The actual water content of the soil was, in both cases, lower than that of test 6 (table 1), although the value of $(a - m)$ was higher in test 7 than in any other test with this soil, which may indicate that since the expression is approximately correct for soils both above and below them in water content, the error is in the determination of soil moisture or soil solids or both. Such circumstances render the loss of more than two-thirds of the data, because of leaking osmometers, regrettable.

Turning to the tests with sandy soil, it will be noted that they exhibited a higher rate than was to be expected from the magnitude of the values obtained for $(a - m)$ (table 1). Test 9, for example, had a total water content only slightly higher than $(a - m)$ for test 7, yet the rates were much greater. The most obvious possibility is that r must not be regarded as a universal constant for all soils, but that it varies in magnitude from one to another. The value of r was accordingly redetermined, the data obtained in test 8 being used. The rate at period 17 was taken from the osmometer data and this, with the corresponding values for the other symbols, was substituted in equation (6).

$$s = 2 + (94/61) (48.1644 - 44.3616) = 7.9$$

$$r' = s/(a - m)^3 = 7.9/11.5 = 0.686$$

When this value of r' is used for test 9, the other test with sand, the value obtained, 20.2, for $r^1 (a - m)^3$ is found to be in good agreement with the value of s , 20.1. This indicates a characteristic difference between the two soils since the value of the constants appears to hold over a considerable range of soil moisture, the entire range for which they have been tested.

The variation in r from one soil to another introduces another obvious

limitation to its use as a constant: it contains no statement of the degree of soil moisture that will produce the highest rate possible. It is apparent that when a certain amount (perhaps characteristic of a given soil) of water is added to a unit volume of soil by displacing contained air, the wedges become completely confluent, the hygroscopic films disappear and the soil mass loses its structure, becoming simply a body of water containing soil grains and gas bubbles. The surface forces no longer operate in the movement of water but are effective only in partially conditioning the form and volume of the gas bubbles. The soil has then passed beyond the range of moisture content for which the proposed equation was formulated, for such a soil is undrained. How sharply this point will manifest itself may only be conjectured. If this point is independent of the percentage of solids in the soil mass, it may be characteristic only of the kind of soil. It would appear that a marked change in specific volume should occur at this point and it may be thus quantitatively the same as the "critical moisture" content of Cameron and Gallagher (11). If these points *are* identical the volume percentage of the contained water should vary with the volume percentage of the solids, as is plainly indicated by these author's observations on wetted and dried soils. The apparent agreement between the "critical moisture" content and the "moisture equivalent" of Briggs and McLane (9) cannot be considered real since the method used by the latter authors (subjection of the sample to great centrifugal force) must result in a packing of the soil so that this "constant" cannot be determined for soils with a low percentage of solids if a constant force is employed.

Since there is undoubtedly a variation from one soil to another in the degree of soil moisture at which surface forces are no longer effective in the movement of water, there is great probability that, of two soils having the same value for $(a - m)^{\frac{1}{2}}$, the one that would have the lowest percentage of water when puddling occurred would exhibit the higher initial rate, for it would be nearer complete saturation than the other. These considerations emphasize the need of further investigations on the "critical moisture" content of soils, especially with regard to a simplification of the method and the relation of this point to that at which puddling occurs.

Another explanation for the relatively smaller rate with the garden soil than with the sand is based upon the decrease in surface tension caused by the fatty substances of the manures that were applied to the former. Whether this had an effect upon the gas-water surface-tension or only upon the solid-water surface tension cannot be stated because of the meagerness of data,⁷ although it seems at present that the latter is the more probable.

In table 1 and in figure 12 are presented the values of s calculated from the observed rates and the value of k , together with the corresponding values of $r(a - m)^{\frac{1}{2}}$. If a straight line is drawn, passing as nearly as possible through all the plotted values of s in the tests with garden soil it intersects the hori-

⁷ Considerable literature on absorption in soils is summarized by Patten and Wagaman (18).

zontal axis at the right of the origin. This may indicate that water may be present in the soil above the α water content and yet permit of no measurable absorption by the osmometer or it may indicate that the curve becomes changed in slope as it approaches the horizontal axis. A line drawn through the two values for s determined from the sand tests, however, cuts the origin almost exactly and in view of the difficulty of obtaining from the present data information regarding the actual values for s uncomplicated by temperature effects and errors of observation, it is idle to attempt a conclusion on this point.

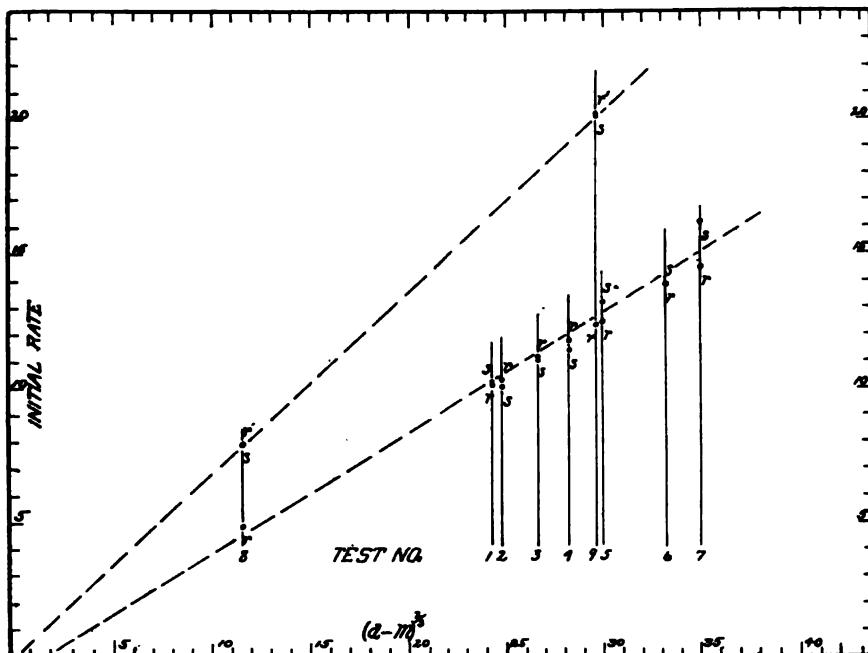


FIG. 12. INITIAL RATES OF THE SOIL TESTS

Points designated r signify the rate was calculated from the expression $r = (a-m)^{\frac{1}{2}}$ in which $r = 0.417$, r' signifies the rate was calculated from $r' = (a-m)^{\frac{1}{2}}$ in which $r' = 0.686$, and s signifies the rate was calculated from equation (6).

3. THE HEIGHTS OF THE GRAPHS DURING THE FIRST FEW MINUTES

Apparently the first few observations in each test are not indicative of the actual movement of water into the osmometer. To obtain a reading truly representative of this movement it is necessary that the instrument, when placed in the soil, shall be at once in full operation and that no change shall occur in the volume of the contained solution, other than that due to the entrance of water. As already noted, complete contact with the soil was assumed only when movement of the osmometer was not followed by an apparent increase in the rate.

The temperatures of the solutions used in the tests were lower than those of the soil in every case except those of 6, 8 and 9. The actual amount of these differences cannot be stated, as some change occurred before the osmometer could be placed in position, but since an approximate change in volume of 10 cu. mm. follows a change in temperature of 1.0°C., the probability of large errors in the first few readings is very great. Although no data are available dealing with the temperatures of the solutions used in the tests with sand it is very probable that the soil was much the colder. The soil temperatures were low and the osmometers and solutions were exposed to direct sunshine during the thirty to forty-five minutes required to transport them to the locality at which the tests were made.

Besides the inaccuracies due to causes already mentioned, the observation of the rate during the first time-interval is probably affected by the drying of the soil surface during the time the osmometer was being placed in position, the excess of water on the membrane, due to incomplete removal with absorbent paper, and deficiency of water in the membrane through exposure to the air. These conditions appear to be only temporary and are, perhaps, of slight importance.

4. THE APPARENT FLUCTUATION IN THE RATES OF WATER MOVEMENT

Of the possibilities in this connection, expansion and contraction of the osmometer solution due to changes in temperature are undoubtedly influential. It is impossible to estimate accurately this class of effects because the rate of temperature change in the solution was necessarily slower than the rate of change in the external temperature, so that the volume of the solution must often have been changing in a direction opposite to that in the external temperature. Since there was some change in the temperature of the solutions the fluctuations in the rates must be ascribed at least to some extent to temperature changes.

The changes in volume of the solution were read on a graduated tube made by cutting off the capillary tip of a Mohr pipette, having a capacity of 1 cc. and graduated to hundredths. Since it was necessary to read to thousandths (cubic millimeters) tenths of intervals were estimated. Errors in estimation of a cubic millimeter were accordingly possible, although the probability of frequent errors greater than this is small.

Fluctuation of the observed rates is also possible through the lack of uniformity in the soil. As has been noted, a soil is in equilibrium when the surface curvatures of all the wedges are the same, regardless of the amount of water contained in the wedges. If the soil were perfectly homogeneous, all grains being of the same shape, touching each other at corresponding points and uniformly distributed, the amounts of water forming the wedges would probably be the same. Without question this condition never obtains. Some wedges, because of accidents of shape and arrangement of soil grains contain

more water than others. Uniformity of distribution of either water or soil grains also certainly never occurs. In consequence of this irregular distribution of water it appears inevitable that the same alteration in capillary tension in different portions of a soil will be accompanied by movement of different amounts of water. In such ways fluctuations in the rate of intake by the osmometers may occur.

Changes in the positions of the soil grains consequent upon water withdrawal would decrease the rate of movement from wedge to wedge and this effect should be most apparent in loosely-packed soils. Reference to the graphs will make evident the apparent fact that, in general, the greatest variation is in such soils, but it should also be noted that it is in such soils that the lack of uniformity in soil and water distribution is the greatest, thus producing the effects discussed in the preceding paragraph.

THE DETERMINATION OF THE PERCENTAGE OF WATER IN THE SOIL ON THE BASIS OF APPARENT, OR GROSS SOIL VOLUME

The usual methods for obtaining the volume percentage of moisture consist of cutting a rather large block of soil, or removing a short column with a tube forced into the soil, or removing a mass of soil with a soil augur. Of the three, the last-mentioned is the best method although it is, like the first, unsatisfactory because small stones and other objects that have no direct effect upon water movement may comprise a part of the soil removed. The first method is also unsatisfactory because the dimensions of the blocks are difficult to determine with accuracy. The second method is of least value since the tube compacts the soil because of friction between the soil mass and the walls of the tube. Compression of the soil in front of the tube will prevent the desired amount of soil from entering and compression within the tube will change the volume of that portion of the soil that is removed, these circumstances preventing any accurate knowledge of the volume occupied by the soil when in its original condition. Packing also occurs when the soil augur is used but to a less extent. The need was accordingly for an instrument with which samples might be removed in practically their original volume. The instrument described below and illustrated in figure 13 was constructed to cut soil blocks quickly and with a high degree of accuracy.

A thin, flat blade of metal was attached to a handle, in such a way as to give a rigid surface about 2 inches broad and 4 inches long. This blade was pointed at the tip and sharpened along the sides and tip to a knife edge. A rectangular box, formed from sheet metal with the upper and lower faces open and with the lower (cutting) edge of the remaining four sides bevelled on the outside (so that all compression of the soil in a direction normal to the direction of movement of the cutting edge was avoided), was hinged to the blade in such a way that it would pivot about an axis parallel to the broad surface of the blade. In operating the instrument, this rectangular box was

turned on its axis until it stood at right angles to the broad blade, which was then forced horizontally into the soil about an inch below the surface. The box was then swung downward to a horizontal position, thus cutting out a small block of the soil that lay above the blade. The entire instrument was then removed, the soil lying outside the box was brushed away, and the soil projecting above the sides of the box was trimmed with a sharp knife to form a surface flush with the sides. The weight of the water and that of the soil contained in the block were then determined in the usual way, by weighing, drying at 103–105°C., and reweighing.

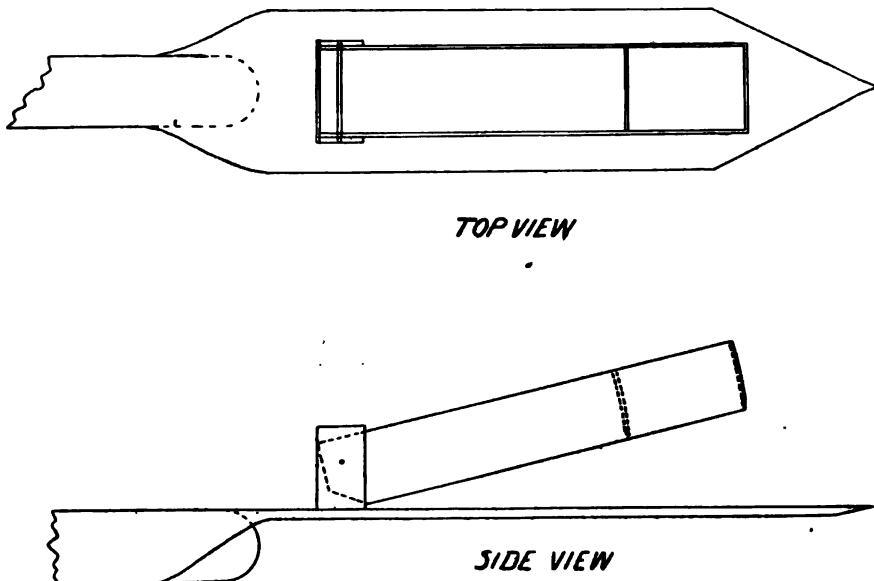


FIG. 13. INSTRUMENT FOR OBTAINING SOIL SAMPLES OF KNOWN FIELD VOLUME

To determine the internal capacity of the box, a block of putty was cut out as described above, removed, and dropped into a graduated cylinder partly full of water. The increase in volume of the contents of the cylinder gave the capacity of the box. Several determinations made in this way indicated that the volume of the soil blocks was 7.6 cc., the greatest variation observed being about 1 per cent.

Soil samples obtained in the field were immediately placed in small glass vials, tightly stoppered with paraffined cork stoppers and weighed indoors to the nearest centigram within five minutes of the time they were obtained. They were later dried at a temperature of 103–105°C. in an electric oven in the same vials with the stoppers removed, cooled in a desiccator over calcium chloride, the stoppers replaced and again weighed.

THE SOILS USED IN THESE TESTS

The garden in which most of the tests were conducted had been terraced up to its present height 10 feet above the land below, so that its level was near the summit of the hill of which it had formerly been the steep eastern slope. It had been in constant use as a vegetable garden for over fifty years and had been almost yearly manured with stable manure. The soil is a very black sandy loam, containing almost no clay, but with considerable amounts of humus. No test was made within 4 feet of the position of any former test.

The soil termed sand formed a 30-foot hill whose steep northern slope extended down to a river. This soil is a fine yellow sand and very uniform. A thin (2 to 3-inch) layer of dark, humus-bearing soil overlay the sand and the osmometers were placed with the upper edge of their membranes about an inch below the junction of the two sorts of soil. Large oak (*Quercus rubra*) elm (*Elmus Americana*) and basswood (*Tilia Americana*) trees grew on this hill in sufficient number to shade the area in which the tests were made. Near the river *Taraxacum officinale*, several ferns, mosses, *Viola*, *Oxalis*, *Cnicus*, *Ribes Cynosbatii* and *Ribes Canadensis* were growing sparsely, and among them test 9 was made. A portion of the slope was much steeper than the remainder, forming a ridge running back from the water's edge. Some distance from the river this ridge supported only infrequent clumps of a coarse grass, and here test 8 was made.

SUMMARY OF FIELD PROCEDURE WITH OSMOMETERS

An osmometer, after being tested for permeability, was filled with M. 1.5 magnesium sulfate solution and carried to the experimental plot (30 to 40 yards in the garden, a few feet when the sand was tested) in water. A wedge-shaped trench was cut in the soil to a depth of about once and a half the height of the osmometer, i.e., about six or seven inches, and slightly narrower at the top than was the osmometer at its widest part. The edges of the trench were trimmed with a sharp knife to a plane surface, making it slightly wider at the top than the maximum diameter of the osmometer. The instrument was then removed from the water, the excess moisture on the surface of the membranes was removed with absorbent paper and it was lowered into the trench until it fitted snugly. Pressure was carefully regulated so that the natural texture of the soil surface was altered as little as possible. The remainder of the trench was filled with the earth that had been removed in its formation, some of which was used to cover the entire osmometer except the horizontal graduated tube. As soon as the instrument was in position an observation was made and further observations were taken regularly during the time the osmometer was in operation. A thermometer was inserted in the soil within an inch or two of the osmometer's position but in an undisturbed portion and another was hung near the first and close to the level of the ground.

TABLE 1
Numerical data for the soil tests
 (Area of membranes 2.863 sq. cm., $r = 0.417$; $r' = 0.686$; $k = 94$; length of period, ten minutes)

NUMBER TEST	SOIL	a (mm) ²	m (mm) ²	$(a-m)$ (mm) ²	$\frac{m}{(a-m)}$ (mm) ⁻¹	$\frac{a}{(a-m)}$ (mm) ⁻¹	$r \cdot \frac{a}{(a-m)}$ (mm) ⁻¹	$r' \cdot \frac{m}{(a-m)}$ (mm) ⁻¹	s	TIME TEST WAS BEGUN	TIME TEST WAS ENDED
1	Garden	193	74	119	24.20	10.1			10.2	9.40 a.m.	2.00 p.m.
2	Garden	200	77	123	24.73	10.3			10.0	12.10 p.m.	8.50 p.m.
3	Garden	213	76	137	26.60	11.1			11.1	10.00 a.m.	7.00 p.m.
4	Garden	211	62	149	28.10	11.7			11.4	10.50 a.m.	7.00 p.m.
5	Garden	237	74	163	29.85	12.5			13.2	3.50 p.m.	8.00 p.m.
6	Garden	260	70	190	33.05	13.8			13.8	11.15 a.m.	5.05 p.m.
7	Garden	257	51	206	34.90	14.6			16.2	10.40 a.m.	4.40 p.m.
8	Sand	100	61	39	11.50	4.8	7.9	7.9	2.50 p.m.	6.00 p.m.	
9	Sand	215	51	164	29.50	12.3	20.2	20.1	11.50 a.m.	3.00 p.m.	

TABLE 2
Numerical data of the test of the osmometer operating against water
 (Length of period five minutes)

PERIOD	cu. mm. AB-SORBED										
1	32	24	31	47	32	70	30	93	31	115	28
2	36	25	31	48	30	71	29	94	28	116	29
3	35	26	31	49	30	72	30	95	27	117	29
4	32	27	33	50	30	73	27	96	27	118	28
5	30	28	31	51	30	74	30	97	30	119	28
6	32	29	30	52	30	75	29	98	29	120	28
7	31	30	33	53	30	76	30	99	27	121	27
8	29	31	31	54	30	77	29	100	31	122	27
9	32	32	31	55	30	78	28	101	29	123	28
10	32	33	30	56	30	79	28	102	*	124	28
11	33	34	*	57	29	80	30	103	27	125	28
12	32	35	32	58	31	81	27	104	29	126	28
13	31	36	27	59	29	82	30	105	29	127	30
14	31	37	31	60	29	83	29	106	29	128	29
15	32	38	33	61	31	84	29	107	28	129	27
16	32	39	31	62	28	85	29	108	29	130	27
17	33	40	30	63	29	86	30	109	29	131	28
18	32	41	30	64	32	87	27	110	28	132	27
19	31	42	30	65	34	88	28	111	28	133	28
20	30	43	29	66	30	89	30	112	30	134	*
21	34	44	32	67	*	90	29	113	28	135	29
22	31	45	29	68	30	91	29	114	27	136	30
23	31	46	30	69	30	92	30				

* Column reset to zero.

On sunny days and during rain the osmometer and thermometers were protected by a canvas shade about 6 feet square.

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EDITORIAL NOTE

Mr. Nicholas Kopeloff, for 18 months assistant editor of *SOIL SCIENCE*, has accepted service with the United States Department of Agriculture and finds it necessary for this reason to sever his connection with *SOIL SCIENCE*. The editors wish to express their feeling of indebtedness to Mr. Kopeloff for faithful and efficient service.

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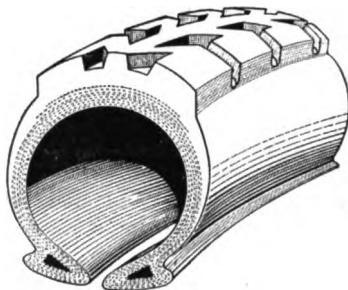


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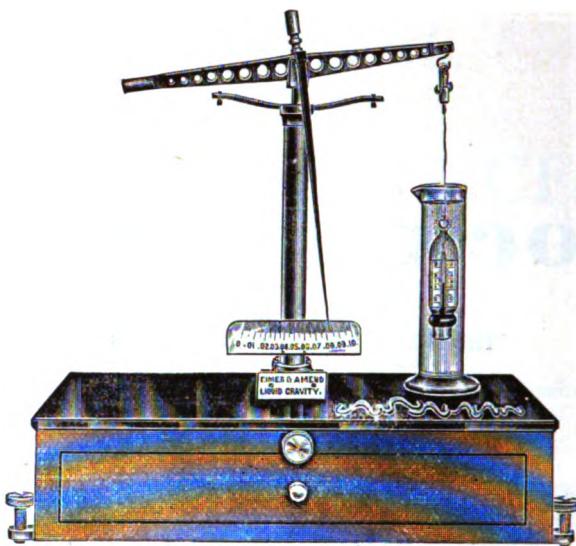
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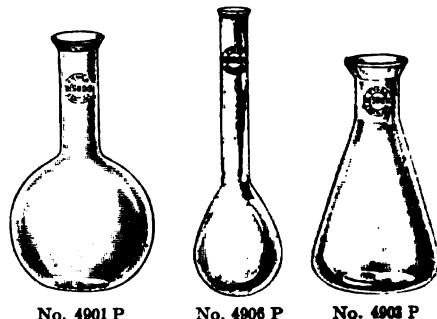
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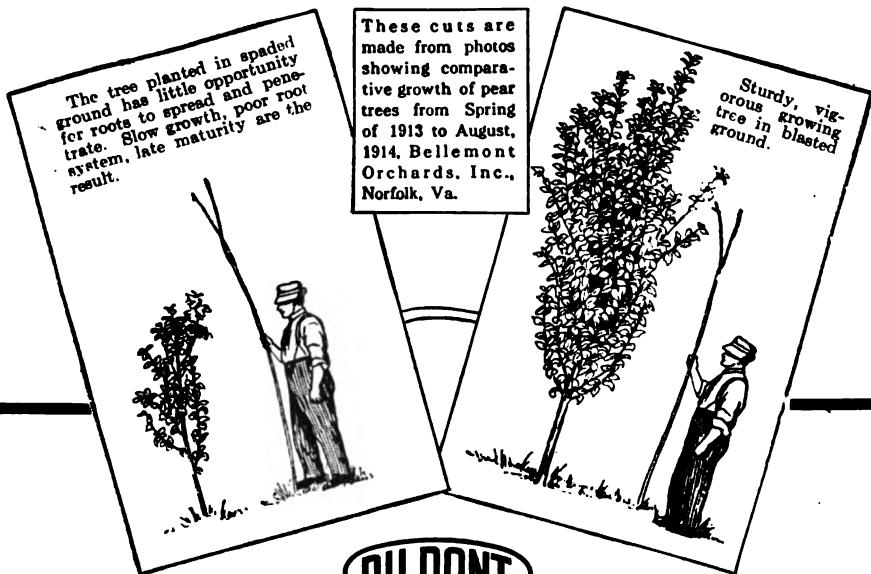
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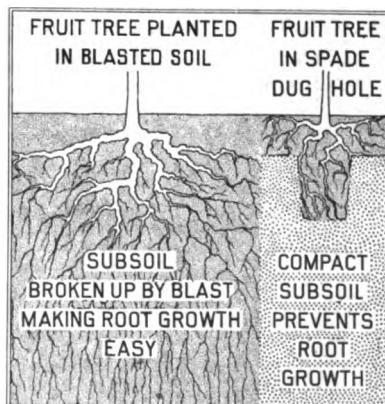
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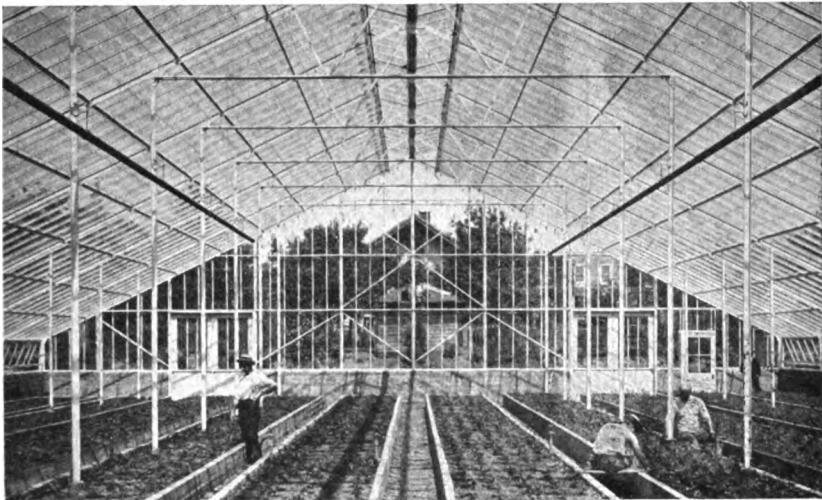
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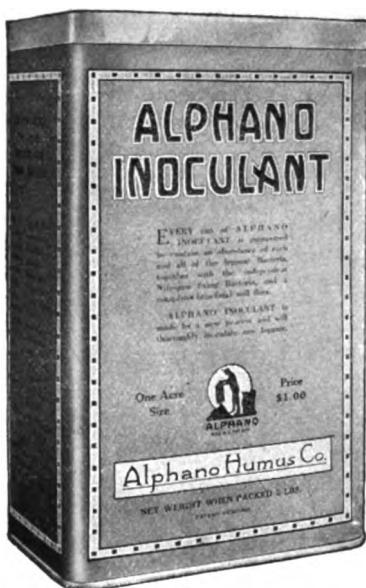
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THE PRODUCTION OF AVAILABLE PHOSPHORUS FROM ROCK PHOSPHATE BY COMPOSTING WITH SULFUR AND MANURE

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Ever since the days of Liebig, the need of the return to the soil of the phosphorus removed by crops has been recognized. Various phosphorus-containing materials have been and are still being employed as fertilizers but the discovery of the extensive deposits of mineral phosphates in various parts of this country has made these materials, at this time, the chief source of phosphorus for general fertilization purposes.

The phosphorus in these mineral phosphates is in an unavailable form and they have been treated commercially with sulfuric acid to produce available phosphorus or "acid phosphate." Considerable expense is involved in this process with the consequent effect that the phosphorus in acid phosphate costs the farmer more than twice as much as that in the raw rock. Attempts have therefore been made from time to time to devise some practical method of treating the raw material on the farm, either before or after application to the soil, to make it available. Many experiments have been carried out, mixing the phosphate with decaying organic matter, but the results have been far from satisfactory. The investigations have been summarized by Lipman, McLean and Lint (5, 6) and the conclusion seems well warranted that this production of acid phosphate on the farm by composting with organic materials is at best a very uncertain process.

The above-mentioned authors suggested the use of free sulfur with rock phosphate to make the phosphorus available. The sulfur is oxidized rapidly in the soil through the action of microorganisms and the sulfuric acid produced reacts with the raw rock, producing acid phosphate in much the same way as the latter material is obtained commercially. This method of producing available phosphorus was tested with several soils and was found to be very efficient in all cases. The soil containing the largest amount of organic matter was found, however, to lead to the greatest increase in available phosphorus. The authors conclude that in field practice, inert phosphates may best be made available by sulfur oxidation in a compost heap containing a relatively large proportion both of phosphate and sulfur.

From a consideration of the investigations briefly referred to above and the conclusions drawn from them, it seemed that acid phosphate might be produced on the farm to good advantage by composting the raw rock with

sulfur and manure. A double purpose might thus be accomplished, not only by producing available phosphorus, but in conserving the manure. Just how efficient such a method of conservation of the valuable portions of manure might be, remains to be ascertained in future studies. The present work is concerned only with the changes which the phosphorus and the sulfur undergo in the manure heap.

Furthermore, if it were possible to produce acid phosphate in the manure heap by composting not only would there be a large saving in the cost of the phosphorus applied to the soil, but also the expense of application would be lessened.

The purpose of the experiments reported in the following pages was therefore to ascertain the feasibility of producing acid phosphate on the farm by composting rock phosphate with sulfur and various farm manures.

EXPERIMENTAL

It was realized that one of the chief factors involved in the success of the process would be the efficiency of the sulfofying flora of the manure. Accordingly, a preliminary series was planned to throw some light on this point and to compare the sulfofying floras of the manures to be tested with that of a rich loam.

One-hundred-gram quantities of an air-dry silt loam were weighed out in tumblers. Each portion of soil received an addition of 100 mgm. of flowers of sulfur, which were thoroughly stirred in. Duplicate portions of the soil were then inoculated with 5 cc. of infusions of compost, horse manure, cow manure and fresh soil, respectively. Water was added in sufficient amounts to bring the content of the soils up to the optimum. The mixtures were incubated for 10 days at room temperature. At the end of that time the sulfates were extracted by shaking with water for 7 hours and determined photometrically according to the method of Brown and Kellogg (2).

The results of this test were as follows:

INFUSION	MGM. OF SULFUR AS SULFATE	AVERAGE
Compost.....	60.80	61.80
	62.80	
Cow manure.....	61.20	62.00
	62.80	
Horse manure.....	62.80	63.60
	64.40	
	62.80	
Loam.....	62.00	62.40

It is apparent from these results that each of the manures in question has a sulfofying flora practically as efficient as that of the rich loam. The horse

manure seemed to give a slightly more vigorous action than the other manures and the soil, but the differences were too small to be distinctive. In every case over 60 per cent of the sulfur added was transformed to sulfates. This would indicate clearly that the manures and the soil tested contain sulfo-fiers of such efficiency and in such abundance as to insure a rapid production of sulfates in the field.

PLAN OF THE EXPERIMENT

The experiment was arranged in three series, a different manure being used in each. Compost, cow manure, and horse manure were used in series A, B and C, respectively.

The compost consisted of a mixture of horse manure, cow manure, straw, hay, and other litter. It represented fairly well the contents of the ordinary manure pile or pit. This material was dried, ground, mixed and analyzed for total phosphorus. The phosphorus content was found to be 0.3232 per cent, on the dry basis.

The horse manure and cow manure were fresh, unfermented materials and contained no straw or litter of any kind. These were treated in the same manner as the compost. The phosphorus content was found to be 0.6051 and 0.5165 per cent, respectively.

The rock phosphate employed was a finely-ground rock, analyzing 12.81 per cent of total phosphorus. Pure precipitated sulfur was used.

Small glass tumblers were used as containers. Therse were fitted with tin covers to prevent excessive evaporation, but loosely enough to permit adequate aeration. In each tumbler was placed 25 gm. of the dry manure, treated in duplicate, as follows:

- Nos. 11 to 15 (inclusive) 25 gm. manure.
- Nos. 21 to 25 (inclusive) 25 gm. manure.
- Nos. 31 to 35 (inclusive) 25 gm. manure + 4 gm. rock phosphate.
- Nos. 41 to 45 (inclusive) 25 gm. manure + 4 gm. rock phosphate
- Nos. 51 to 55 (inclusive) 25 gm. manure + 1½ gm. sulfur.
- Nos. 61 to 65 (inclusive) 25 gm. manure + 1½ gm. sulfur.
- Nos. 71 to 75 (inclusive) 25 gm. manure + 1½ gm. S. + 4 gm. rock phosphate
- Nos. 81 to 85 (inclusive) 25 gr. manure + 1½ gm. S. + 4 gm. rock phosphate.

Each series was treated the same, the only difference being in the manure used. All additions were thoroughly mixed with the manure and the contents of each tumbler were inoculated with 2 cc. of a corresponding infusion of fresh manure. Sufficient water was added to bring the contents of the soils up to the optimum and they were incubated at room temperatures, the water lost by evaporation being restored every three weeks.

The entire contents of the pots were taken for analysis. This was done in order to eliminate the possibility of not securing representative portions of the mixtures. Determinations of available phosphorus were made at the following intervals:

- Nos. 11, 21, 31, 41, at beginning (no fermentation).
 Nos. 12, 22, 32, 42, after 6 weeks' fermentation.
 Nos. 13, 23, 33, 43, after 9 weeks' fermentation.
 Nos. 14, 24, 34, 44, after 12 weeks' fermentation.
 Nos. 15, 25, 35, 45, after 15 weeks' fermentation.

Available phosphorus determinations

The methods now in use for determining available phosphorus are quite arbitrary, and in no way can they be considered even as approximations of the phosphorus actually available for plant use. In this work, the phosphorous was extracted by N/5 HNO₃ with 5 hours' constant shaking at room temperature. The N/5 HNO₃ is perhaps a more active solvent than some of the other extraction-solutions, but it undoubtedly gives dependable comparative results.

Fraps (4), Tottingham and Hoffman (7) and Ames and Gaither (1) have used this acid for phosphorus extraction. The greater convenience of preparing and working with N/5 HNO₃, as compared with some of the other solvents, was also considered in the selection of this solution as the measure of availability.

To determine the available phosphorus by this method the contents of the tumblers were transferred to shaker bottles with 250 cc. of N/5 HNO₃. These were shaken for 5 hours in a mechanical shaker. The contents were filtered and duplicate 50-cc. portions of the filtrate were evaporated to dryness with 5 cc. of Mg(NO₃)₂. The residues were ignited and dissolved in HNO₃ and a little HCl. The phosphorus was then precipitated in the usual way and determined volumetrically.

Series A

In table 1 are given the results of the determinations made in Series A where compost was used as the manure with which the floats and sulfur were composted.

TABLE 1
Determinations of available phosphorus when compost was used

	COMPOST mgm.	COMPOST AND SULFUR mgm.	INCREASE DUE TO SULFUR mgm.	COMPOST AND FLOATS mgm.	COMPOST, SULFUR AND FLOATS mgm.	INCREASE DUE TO SULFUR mgm.
Total Phosphorus.....	80.8	80.8		593.2	593.2	
Time:						
At beginning.....	49.3	49.3	0.0	289.9	289.9	0.0
End of 6 weeks.....	47.9	56.9	9.0	249.5	342.2	92.7
End of 9 weeks.....	58.3	63.3	5.0	255.5	371.0	115.5
End of 12 weeks.....	59.9	63.3	3.4	268.9	395.4	126.5
End of 15 weeks.....	57.6	60.9	3.3	272.9	428.6	155.7

It is evident from these figures that in the untreated compost, fermentation caused a rather noticeable increase in available phosphorus which apparently reached its maximum at the end of 9 weeks, the variations after that time being too small to be distinct. The compost treated with sulfur gave a similar increase, which was slightly greater than where no sulfur was added. The addition of sulfur apparently enhanced the solubility of the phosphorus although to a rather limited extent. When rock phosphate, containing

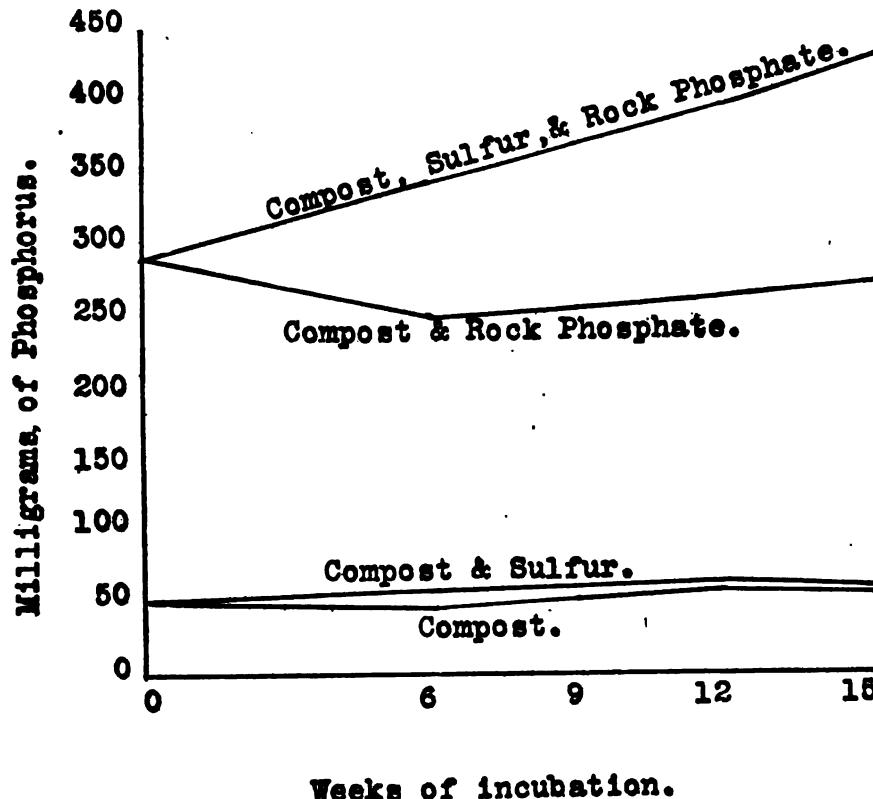


FIG. 1. DIAGRAM SHOWING DETERMINATIONS OF AVAILABLE PHOSPHORUS WHEN COMPOST WAS USED

512.4 mgm. of phosphorus, was added to the manure, there occurred at the beginning, an enormous increase in available phosphorus. This increase represented 45 per cent of the total phosphorus in the mixture. Evidently the solution used for extraction dissolved much more phosphorus from the floats than would be readily available to plants. Six weeks of fermentation caused a very noticeable depression in phosphorus-availability. After that time, however, there was a continuous, gradual increase, up to 15 weeks, but at that time the original degree of solubility had not quite been attained.

With longer incubation, further increases in available phosphorus might have occurred and the amount produced might have been far in excess of that obtained at the beginning.

Where sulfur was present with the compost and floats an unquestionable increase in availability was found. With only 6 weeks' composting, the sulfur caused a greater solubility of phosphorus by 92.7 mgm., as compared with the mixtures containing no sulfur. The increases became greater at each sampling until at the final date a gain of 155.7 mgm. of phosphorus, or 26 per cent of the total phosphorus present, was obtained, due to the action of the sulfur. The addition of sulfur to the floats and manure evidently not only offset the depression brought about in the mixture of compost and floats, but caused a very marked gain in the solubility of the phosphorus in the floats. The results obtained in this series furnish very definite evidence in support of the theory that the phosphorus in floats may be made available by composting with manure and sulfur. The results of Series A, as given in table 1, are shown graphically in figure 1.

Series B

The results given in table 2 show the amounts of available phosphorus found at the various intervals in mixtures of cow manure with floats and sulfur.

TABLE 2
Determinations of available phosphorus when cow manure was used

	COW MANURE	COW MA- NURE AND SULFUR	INCREASE DUE TO SULFUR	COW MA- NURE AND FLOATS	COW MA- NURE, SULFUR AND FLOATS	INCREASE DUE TO SULFUR
	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
Total Phosphorus.....	129.1	129.1		645.1	645.1	
Time:						
At beginning.....	103.9	103.9	0.0	307.9	307.9	0.0
End of 6 weeks.....	79.5	100.3	20.8	284.9	421.5	136.6
End of 9 weeks.....	80.6	96.6	16.0	297.7	437.9	140.2
End of 12 weeks.....	81.6	97.6	16.0	285.9	444.5	158.6
End of 15 weeks.....	83.3	99.3	16.0	279.9	464.2	184.3

It is evident from these data that the availability of the phosphorus in the untreated cow manure was greatly lessened by fermenting for 6 weeks. This is in direct opposition to the corresponding results in the compost series, where an increase in availability was found. After 6 weeks there was a gradual increase in the availability which was so small as to be of no consequence. This decrease in available phosphorus is probably the result of the vigorous action of phosphate-assimilating organisms in the early stages of fermentation in fresh manure.

Where sulfur was added to the manure a very slight decrease in available phosphorus occurred. Here the presence of sulfur practically prevented

any depression in phosphorus-solubility, but did not cause any greater solubility from the original analysis to later ones. Of the 512.4 mgm. of phosphorus in the rock phosphate added to the manure, 204.0 mgm. were found to be soluble, at the first sampling. At later dates smaller amounts of available phosphorus were obtained, the decrease becoming slightly greater at the end of 15 weeks. If the experiment had been continued for a longer

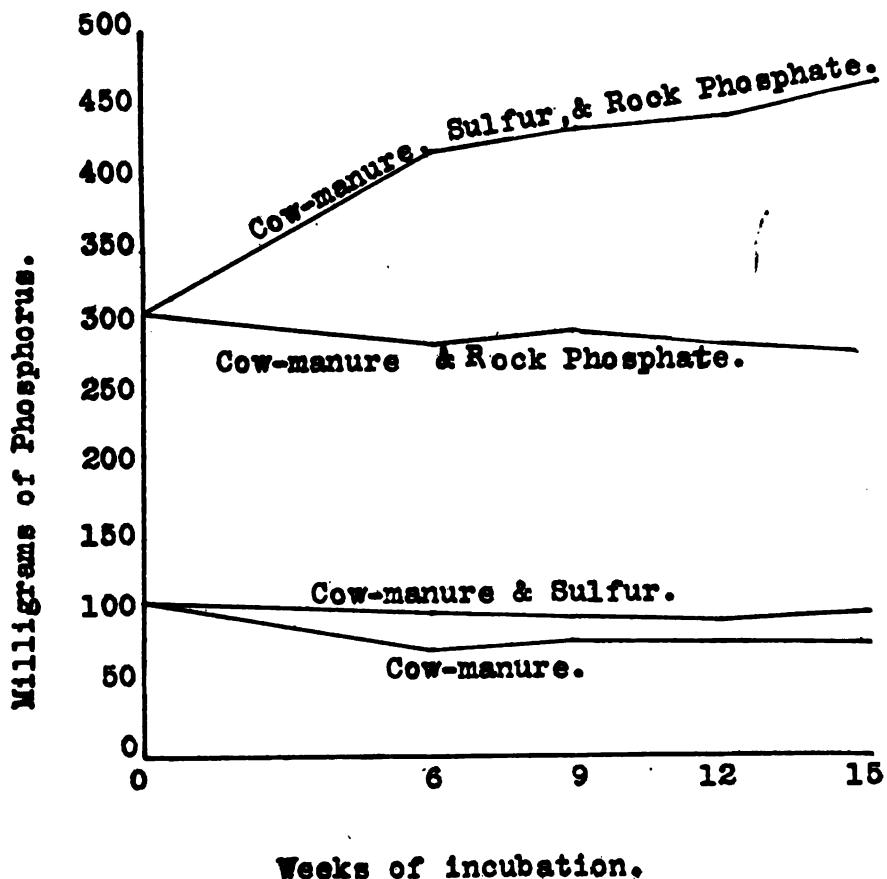


FIG. 2. DIAGRAM SHOWING DETERMINATIONS OF AVAILABLE PHOSPHORUS WHEN COW MANURE WAS USED

time an increase in available phosphorus might have been secured. Evidently the activities of phosphate-assimilating organisms is great enough in the early stages of fermentation of manure to reduce the availability not only of the phosphorus in the manure but of that added in the phosphate.

Where sulfur had been added with the floats the effects on the production of available phosphorus are quite distinct. After 6 weeks of composting, the sulfur produced the remarkable increase of 136.6 mgm. of available

phosphorus over the manure-floats mixtures. Larger increases were found at each succeeding date and the greatest effect of the sulfur was found at the final analysis when 184.3 mgm. of phosphorous were made soluble. This amount equalled 29 per cent of the total phosphorus in the manure and floats. As in the preceding series the action of the sulfur not only overcame the depression in phosphorus-availability in the manure-floats mixture but also brought about a very marked increase in soluble phosphorus at the successive periods of composting. Series B affords additional data confirming the theory that the phosphorus in floats may be made available by composting with manure and sulfur. Figure 2 shows graphically the numerical results expressed in table 2.

Series C

In this series horse manure was used as the medium to which the floats and sulfur were added. The results of the determinations for available phosphorus are recorded in table 3.

TABLE 3
Determinations of available phosphorus when horse manure was used

	HORSE MANURE	HORSE MA- NURE AND SULFUR	INCREASE DUE TO SULFUR	HORSE MA- NURE AND FLOATS	HORSE MA- NURE, SULFUR, AND FLOATS	INCREASE DUE TO SULFUR
	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
Total Phosphorus.....	150.8	150.8		663.2	663.2	
Time:						
At beginning.....	107.9	107.9	00.0	363.9	363.9	00.0
End of 6 weeks.....	106.8	110.9	4.1	342.9	415.2	72.3
End of 9 weeks.....	102.3	113.6	11.3	338.2	433.9	95.7
End of 12 weeks.....	89.5	113.6	24.1	350.9	451.5	100.6
End of 15 weeks.....	99.9	112.3	12.4	362.5	481.9	119.4

As in Series B, there was a depression in available phosphorus in the untreated manure. This was greatest at the end of 12 weeks, the final analysis exhibiting a distinct increase in the phosphorus extracted, but the amount was less than that secured in the unfermented manure. Upon addition of sulfur, this depression was eliminated and actual increases over the original analysis are to be noticed. The effect of the sulfur was greatest at the 12 weeks' sampling when its presence caused an increase of 24.1 mgm. of available phosphorus over the manure to which no sulfur had been added.

Of the 512.4 mgm. of phosphorus in the floats added to the manure, 256.0 mgm. are indicated as available, at the first analysis. A decided decrease in available phosphorus then took place until after the 12 weeks' interval, when an increase occurred. The final extraction of the manure-floats mixture was only very slightly less than the original extraction of the same mixture. These results are similar to those secured with the compost and the reverse of those with the cow manure.

Again, the addition of sulfur to the manure and floats appreciably enhanced the solubility of the phosphorus in addition to overcoming the lessened solubility found in the manure-floats mixtures. At the end of the experiment there was an increased availability of 119.4 mgm. of phosphorus, or 18 per cent of the total, which increase was due to the action of the sulfur added.

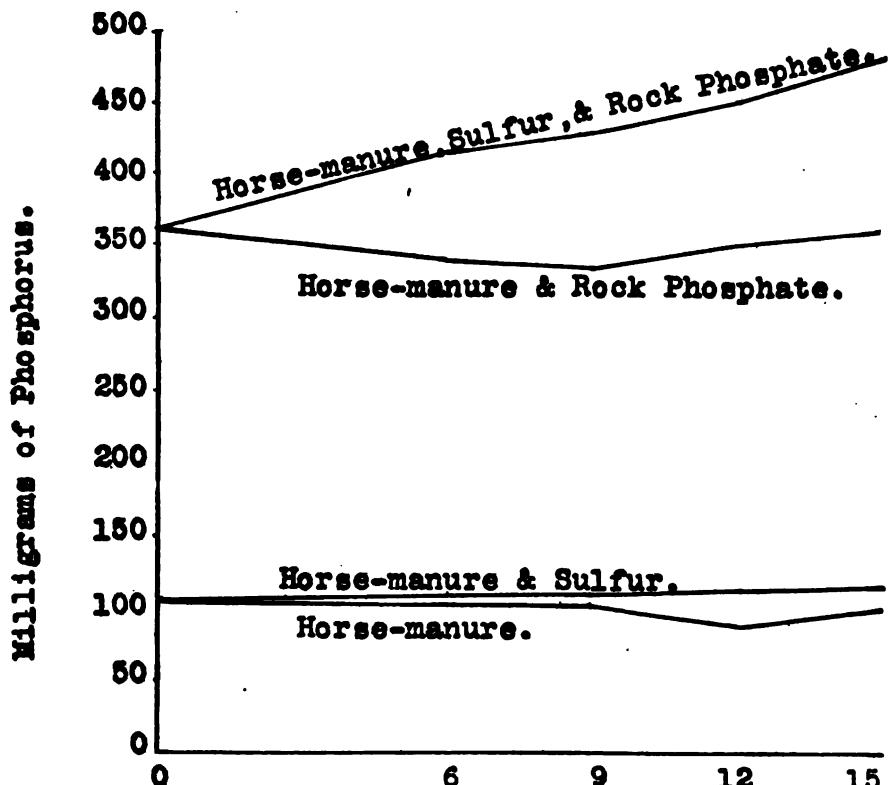


FIG. 3. DIAGRAM SHOWING DETERMINATIONS OF AVAILABLE PHOSPHORUS WHEN HORSE MANURE WAS USED

The results of this series check those of series A and B in substantiating the theory upon which the experiment was based. Figure 3 is the graphic representation of the results tabulated in table 3.

DISCUSSION

For the most part, a rather close agreement is found between the results of these three series. The most noticeable exception to this occurred in the case of the untreated manures, where the fermentation increased the available phosphorus in the compost material but decreased it in both the cow manure and horse manure. The cause of these differences is uncertain but

it is probable that the variations in the physical, chemical and bacteriological conditions of the various manures had much to do in bringing about these results. The depressions found in the amounts of available phosphorus in the horse manure and cow manure agree with the results obtained by Egorov (3). When sulfur was added to the manures there was a more or less apparent gain in available phosphorus, as compared with the untreated manures. In the horse manure-sulfur mixtures and compost-sulfur mixtures, the final results were somewhat higher than the original results. The reverse was true in the cow manure-sulfur mixtures although the difference between the first and last analyses, in this series, was inconsiderable.

Undoubtedly, the method of extracting the available phosphorus indicated a greater amount of phosphorus than would be readily available of plants. Where 512.5 mgm. of phosphorus were added to the manures, increases were found of 240.6 mgm., 204 mgm. and 256 mgm. of soluble phosphorus, in Series A, B and C, respectively, before fermentation. This shows, further, that when the floats were mixed with the various kinds of manures, different amounts of phosphorus were extracted. It would seem that the manures absorbed different quantities of the extracting-acid, thus leaving unequal amounts of this solution free to act upon the particles of floats. It is possible, too, that one kind of manure may have offered more physical protection to the floats particles than did another kind. Although N/5 HNO₃ probably yielded high absolute amounts of available phosphorus, it undoubtedly yielded reliable comparative amounts.

In each of the three series, fermentation depressed the solubility of the phosphorus in the manure-floats mixtures. In series C, the final analysis of the manure and floats showed almost the same availability as the first analysis. This would indicate that, after a period of depression, there is a tendency to regain the original solubility. The same phenomenon was found to occur in the compost-floats mixtures. The depression of available phosphorus in the manure and floats is probably attributable to bacterial assimilation of the dissolved phosphorus. The cause of the increases subsequent to the decreases may be that after a certain time the phosphorus-assimilating bacteria cease to develop proportionately to the phosphorus-liberating bacteria, which condition then permits of the accumulation of soluble phosphorus. It is of interest to note that in no case did composting of floats with manure result in increased availability of the phosphoric acid. This agrees well with the results of several other investigations.

The enormous gains in available phosphorus where sulfur was added with the rock phosphate are the significant results of this experiment. These gains in phosphorus-solubility are so consistent and of such magnitude that the effect of the sulfur can hardly be questioned. After 15 weeks of composting the available phosphorus, as a result of the action of the sulfur, amounted to about 24 per cent of the total phosphorus in the mixtures of manure-sulfur-floats in every series. The constant increases in phosphorus-availability

with the successive analyses indicate that, upon longer composting, a greater percentage of the floats would be made available. It is very evident that the sulfur was oxidized to sulfuric acid by the sulfofying organisms introduced in the manure infusions. The sulfuric acid thus formed reacted with the insoluble rock phosphate and produced acid phosphate in the same way that this product is made commercially.

It is a common practice to apply manure with rock phosphate with the idea of making the latter available in the soil. It should be equally practicable to apply a mixture of these materials with sulfur after composting, and especially if the phosphorus can be applied in a more available form.

There are several considerations which must be taken into account in the application of this composting process to farm practice. Some of these are: the ratio of sulfur to the floats and the proportion of these materials to the manure in the composting mixture; the length of time for a maximum production of available phosphorus in the compost; and finally, and most important of all, the economic consideration.

Commercially, acid phosphate is prepared by treating rock phosphate with an equal part by weight of 60 per cent sulfuric acid. About 400 pounds of sulfur are contained in 1 ton of this acid, hence the sulfur and floats should probably be mixed in the proportion of about 1 part of sulfur to 5 parts of floats. The amount of this mixture to use per ton of manure must be governed by the amount of manure and the amount of rock phosphate to be applied. Experiments along this line are desirable, for while there might probably be no fixed mixture which would be the most profitable, there would undoubtedly be a minimum amount of manure which should be used with a certain amount of rock phosphate.

The results of this experiment show that the longest time of composting gave the highest availability of the phosphorus. Ordinarily it would be possible and practicable to allow the late summer, fall and winter manures to compost until spring. This would permit of from 2 to 4 months' composting in which time a sufficient amount of available phosphorus would be produced to meet the early requirements of the crop. The continued action after applying the composted mixture to the soil would make an adequate accumulation of available phosphorus for the later growth of the crop.

To know whether it is economical to produce available phosphorus by this method it must be tested under actual field and crop conditions. Some theoretical estimations can be made, however, which should indicate rather definitely the economy of such a practice. Since the "compost" material used in this experiment most nearly represented the mixed barnyard manures, the calculations should be based on the results in series A. The 25-gm. portion of dry compost was equivalent to 100 gm. of the fresh material. Hence, in the experiment, the floats were added in the proportion of 4 to 100 or 80 pounds of floats per ton of fresh manure. The floats contained 12.81 per cent of phosphorus. Thus 10.25 pounds of the element phosphorus

were added in each 80-pound portion of raw rock. After 15 weeks there was an increased availability equal to 26 per cent of the total phosphorus in the mixture. The total phosphorus in one ton of fresh manure and 80 pounds of floats was 11.85 pounds. Therefore, an increase of 26 per cent of the total phosphorus would be equivalent to 3.08 pounds of available phosphorus per ton of the mixture. Assuming that the N/5 HNO₃ extracted twice as much phosphorus from the mixture as would be readily available to plants, the amount of available phosphorus produced would be reduced to 1.54 pounds. Then, in a 10-ton application of this composted mixture containing 800 pounds of rock phosphate, there would be, presumably, 15.4 pounds of available phosphorus produced in 15 weeks' fermentation. This amount alone is enough for a 90-bushel crop of corn or a 60-bushel crop of wheat. But the production of available phosphorus would not cease upon application of the mixture to the soil. It has been shown that the production of soluble phosphorus by sulfur-oxidization goes on very rapidly in a soil medium. Hence the conclusion seems justified that there would be a continuous action going on in the soil until practically all of the rock phosphate became converted to acid phosphate.

Theoretically, the foregoing discussion and estimates show that a sufficient amount of available phosphorus for crop requirements can be produced by the proposed method, and that the entire application of floats can be made available upon further action in the soil. Rock phosphate costs about \$10.00 a ton, including freight, and 400 pounds of sulfur cost about \$8.00. Thus the cash investment necessary for making a ton of floats available would be about \$18.00. On the basis of 12.81 per cent of phosphorus in rock phosphate, it would be possible to make 256 pounds of available phosphorus at a cost of \$18.00, or 7.0 cents per pound of phosphorus. Commercially manufactured acid phosphate costs about 15 cents per pound of phosphorus contained.

From the estimates made above and from the calculations based upon these estimates, it appears highly probable that rock phosphate can be made available in a practical and economical way on the farm. The proposed method is based upon sound bacteriological and chemical principles and is in no way impracticable in its application. Actual field tests must of course be carried out before definite recommendations can be made as to the advisability of adopting this process on the farm.

Series D

As the foregoing experiments indicate rather definitely the possibility of producing available phosphorus by composting rock phosphate with sulfur and manure, it was deemed advisable to determine the effect of thorough mixing or layering of the floats and sulfur with the manure.

Accordingly, a series was planned in which different amounts of rock phosphate and sulfur were added to horse manure, the additions being thor-

oughly mixed in one half of the tumblers and layered in the other half. Rock phosphate was used in 1-, 2-, and 3-gm. quantities with the same proportionate amount of sulfur as was employed in the other series— $1\frac{1}{2}$ to 4. All the tests were made in duplicate. The methods used for inoculation, incubation, sampling, analysis, etc., were the same as described for series A, B and C. Analyses for available phosphorus were made at the beginning, at the end of 3 weeks, and at the end of 6 weeks.

Table 4 shows the arrangement of this series with the results of the determinations of available phosphorus.

TABLE 4

Determinations of available phosphorus when different amounts of rock phosphate and sulfur were used with horse manure

TREATMENT			AVAILABLE PHOSPHORUS		
Manure	Floats	How added	At the beginning	After 3 weeks	After 6 weeks
gm.	gm.		mgm.	mgm.	mgm.
25	1	Mixed	188.0	170.7	190.3
25	1	Layered	188.0	170.3	179.5
25	2	Mixed	286.1	271.4	292.7
25	2	Layered	286.1	264.1	278.5
25	3	Mixed	337.2	360.5	389.8
25	3	Layered	337.2	329.7	352.5

Considering the results in this table, it is apparent that the extraction made before fermentation showed nothing of consequence, except that the amount of phosphorus extracted was inversely proportional to the amount of floats added, per gram of the floats used. For instance, where 1 gm. of floats was added to the manure, there were extracted 188.0 mgm. of phosphorus. One additional gram of floats gave an increase of 98.1 mgm. of phosphorus. The next additional gram of floats gave an increase of only 51.1 mgm. of phosphorus. Three weeks of fermentation resulted in a decrease in phosphorus-availability in every case, except where 3 gm. of the floats were mixed with the manure. At this time, the advantage of mixing as compared with layering was quite evident, particularly where the larger amounts of rock phosphate were added. The depressions noted were not entirely overcome at the end of 6 weeks. Only where 1 and 2 gm. of floats were layered with the manure did this depression remain at the last sampling. Evidently, small amounts of phosphate mixed with manure and sulfur are not rendered available as rapidly as the larger amounts. The ratio of 3 gm. to 25 gm. of dry manure seems to be the smallest proportion which allows the sulfur to increase the production of available phosphorus.

The final sampling showed a very noticeable gain in availability where the additions were intimately mixed, as compared with the pots in which the additions were layered. In every case, the results of the experiment show

rather distinctly the advantage of thoroughly mixing over layering when sulfur and floats are composted with horse manure. There is evidently also the need of tests to show the proportion of phosphorus and sulfur which should be mixed with manure.

CONCLUSIONS

The results of these experiments would seem to justify the following conclusions.

1. All of the manures and the loam tested contained efficient sulfofying organisms. The variations in the efficiency of the organisms in the manures and in the soil were too slight to be distinctive.

2. A depression in phosphorus-availability followed the fermenting of the untreated horse manure and cow manure, but in the compost a slight increase occurred.

3. The addition of sulfur to the manures resulted in greater solubility of the phosphorus than was found in the untreated manures.

4. Composting floats with manure not only resulted in no increased availability of the phosphorus but in every case caused a noticeable depression which was not overcome at the end of 15 weeks' fermentation.

5. Composting floats with manure and sulfur caused a remarkable increase in the production of available phosphorus which became greater with longer continued fermentation up to 15 weeks. This increase was greater where the sulfur and floats were intimately mixed with the manure than where they were added to the manure in layers.

6. Experiments are necessary to ascertain the best amount of phosphate to mix with sulfur and manure to secure the highest percentage of availability.

7. Further tests of this method of producing available phosphorus may prove it to be of considerable practical value.

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TOTAL NITROGEN AND CARBON IN CULTIVATED LAND AND LAND ABANDONED TO GRASS AND WEEDS

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INTRODUCTION

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A deficiency of nitrogen in the soil so often becomes a limiting factor in crop production that any investigation which has as its object a determination of the cause of losses, or methods of increasing the supply, of this element, is of prime importance.

It is already well known that under continuous cropping nitrogen does disappear rapidly from cultivated soils, and that frequently the supply is not maintained even when fair amounts of nitrogenous fertilizers are applied annually (6). That the amount removed in the crop does not account for the total loss is likewise well known, but beyond the fact that a part of it is found in the drainage waters as nitrates, the information is not very definite.

In this connection Snyder (14) discussing the effect of the continuous growing of wheat on the same land for a period of years says:

When wheat was grown continuously upon the same soil for eight years, there was a loss of 1700 pounds per acre of nitrogen, about 300 pounds being utilized as plant-food and 1400 pounds lost by the decay of the animal and vegetable matter of the soil and the liberation of the nitrogen as gaseous and soluble compounds. During the eight years of continuous wheat cultivation there was a loss of over 21 per cent of the total nitrogen of the soil, equivalent to an annual loss of 175 pounds per acre in addition to that used as plant-food.

He found about the same loss when oats and barley were grown continuously. The same author in a later publication says: (15)

The loss of nitrogen from four grain farms in ten years amounted to three to five times more than was removed by the crops. This loss was due to the rapid decay of the humus and the liberation of the nitrogen, which forms an essential part of the humus. When clover was grown, crops rotated, live stock kept and farm manure used, an equilibrium as to the nitrogen content of the soil was maintained, the mineral plant-food was kept in the most available condition and maximum yields were secured.

Alway, who has made an exhaustive study of the loess soils of Nebraska, says (1): "Analyses show a great loss of nitrogen, humus and unhumified organic matter in the case of all fields long under cultivation." In their virgin condition he finds these soils rich in the constituents mentioned.

Under a consideration of the nitrogen problem Russell (11) says:

No element of plant-food is so liable to waste as nitrogen. If a farmer applies potash or phosphoric acid to his land he may reasonably expect to get most of it back again, but when he applies nitrogen he is almost certain to lose a good proportion of it. The sound practice is therefore to keep the nitrogen circulating, to crop it out and replace it, rather than to aim at leaving it in the soil.

Shutt (13) calls attention to the unusually high nitrogen content of some of the soils of Manitoba and Saskatchewan (0.2 per cent to 1.00 per cent) but points to the fact that the system of grain growing now in practice on some of the lands results in a large loss of nitrogen annually.

On the other hand, it has been claimed that land allowed to run wild increases in fertility. In this connection Hall (4) calls attention to the fact that a piece of land at Rothamsted which was allowed to run wild contained, at the end of 20 years, 2200 pounds more of nitrogen than it contained at the beginning of the period. This means an increase of more than 100 pounds per acre annually. Hall thinks this increase may be due to symbiotic and non-symbiotic fixation, absorption of ammonia, rise of nitrates from the lower subsoil, and nitrogen compounds brought down in rain water.

Another (2) writer claims that plants derive much more plant-food from soils than chemical analysis accounts for. He says:

A large amount of incidental plant-food, is evidently conveyed by rain water. It appears probable also that the fertility given by the deposited excreta of small animals, birds, insects and worms and reptiles, and the decay of their bodies upon the ground and also the decay of vegetable matter, dust deposited by the wind, leaves, flowers and seeds and branches, etc., is largely in excess of what has previously been estimated, and in fact it is of as much importance as food obtained from the reserves held by the soil itself. This must be known as the incidental increment and must be taken into full account in all agricultural operations.

Hall (5) in a later publication calls attention to the fact that

with every system of farming a certain position of equilibrium will be reached (viewed over a term of years long enough to smooth out seasonal effects) when the natural recuperative agencies and the additions of fertilizing materials in the manure are balanced by the removals in crops and stock and the inevitable waste.

The higher the level of production, the greater will be the waste, and in consequence, the addition of fertilizer must be doubly increased to maintain the balance.

Schneidewind (12) conducted experiments which gave indication that under ordinary methods of intensive culture with liberal use of manure and fertilizers there was a marked depletion of the nitrogen and potassium supply of the soil. There was a relative increase of the phosphoric acid under such a system. Perotti (10) discussing gain of nitrogen, considers bacteria, molds, algae and legumes; and under loss, volatilization of ammonia and denitrification. Shutt (13), by analyses of virgin and cultivated soils, has called attention to the exhaustion of soils by continuous cropping without manure and has shown by experiment that a soil successively cropped to clover continues to increase in nitrogen under this treatment.

Moores, Hampton and Hunter (9) have shown that land in sod for a number of years, suffered a serious loss of nitrogen during five years' cultivation with a double crop rotation of cowpeas and wheat. In this connection they say:

The fact, therefore, that the soil content of nitrogen was not even maintained where a leguminous green crop equal to more than a ton per acre of thoroughly cured hay, on the average, not to mention the fallen leaves, stubble and roots, was turned under each year is almost startling and shows the necessity of a superior system of management if anything like high fertility of the soil is to be maintained without recourse to outside supplies of nitrogen, such as would be furnished by nitrogenous fertilizers and commercial feeding stuffs.

In summing up their conclusions they say further:

By taking the nitrogen removed by the crop into consideration, there was found to be, at the end of five years, (1) where the cowpea crop was turned under, a gain per acre on the unlimed sections of 67 pounds, to be accounted for in sources outside of the 8-inch depth of plowing, but on the limed sections none; (2) where the cowpea crop was removed, a gain per acre of 181 pounds on the unlimed and 177 pounds on the limed, to be accounted for as before.

Swanson (18) states that the soils of Kansas show an average decrease in crop-producing power in spite of the fact that farmers use good seed and improved methods of tillage. Results based on the analysis of cultivated and uncultivated soils in seven representative counties, show that carbon and nitrogen have disappeared from the cultivated to a greater extent than from the uncultivated soils. In round numbers they have lost from one-fifth to two-fifths of their nitrogen and from nearly one-fourth to one-half of the carbon.

Somerville (16) in making a study of the effect of applications of phosphatic manures (notably basic slag) took portions of soil from treated and untreated plots that have been under observation in different sections of England for the past 20 to 30 years and with this soil conducted pot experiments using oats as the first crop. He reports notable increases in yield from the treated soil over the untreated soil. He says:

While it would appear, that the accumulated fertility is partly due to phosphate residues, it is certainly due in most part to the secondary effects of the phosphates, of which the accumulating of humus, including the accumulation of atmospheric nitrogen, is probably the most important.

In a second article (17) he attributes the accumulated fertility largely to nitrogen stored up by leguminous plants, but believes the increase in non-leguminous humus is not without influence.

In an article on the chemical composition of virgin and cropped Indiana soils, Connor (3) says:

In spite of the fact that the tendency of nature is to build up and replenish the fertility of the soil, there is no question but that the destructive system of cultivation that has been followed by the farmers of this country has more than counter-balanced nature's tendency to buildup, and as a consequence the soil has become more or less depleted.

In this connection Connor reports analyses of a number of virgin and cropped soils, and these analyses, he believes, indicate certain changes in the chemical composition of the soil which sixty to eighty years of cropping have brought about. He finds:

that although most of the soil ingredients have not changed enough to make any great difference in the chemical composition of the virgin and cropped soils, there are some notable exceptions.

The most serious losses from the standpoint of soil fertility are those of nitrogen, which shows a loss of 28 per cent, and the organic matter which shows a loss in the volatile matter of 26 per cent and in the humus of 47 per cent. These losses are without doubt the main reason why our cropped soils are no longer as fertile as they formerly were.

If one may judge by the references cited there seems little room to doubt that under continued cropping soils are rather rapidly depleted of their nitrogen and organic matter, and that there may be an actual loss of nitrogen and carbon even when leguminous crops are turned under and manures applied. With uncultivated soils, on the other hand, the losses are not so great and in some instances there are pronounced gains.

EXPERIMENTAL

One of the objects of this experiment was to study the changes which occur in the vegetation on abandoned land and especially the effect of commercial fertilizers on the appearance or disappearance of certain species of plants on such land. The experiment also includes a study of the composition of the soil on the abandoned land as compared with the composition of soil from adjoining plots which are under a 5-year rotation of corn, oats, wheat and two years of grass (originally two years of oats), and the determination of the percentage of applied nitrogen that is recovered in the crop. The soil is a loam inclining to the gravelly phase, with a sandy loam subsoil. Previous to 1908 this land had been neglected for a number of years. The plots were laid out one-twentieth acre in size, one-half to be under cultivation and the other half to remain uncultivated. Beginning with the corn crop of 1908 the plots received a dressing of ground limestone at the rate of 1 ton per acre and beginning with the second five-year period in the spring of 1913 they received a further application of this material at the rate of 2 tons per acre. The uncultivated halves have received the same fertilizer and lime treatment as the cultivated halves. The plots are arranged in pairs with reference to the fertilizer treatment as indicated in the accompanying diagram.

The nitrogen is supplied in the form of ground fish at the rate of 600 pounds per acre, the phosphoric acid as acid phosphate at the rate of 600 pounds per acre and the potash as muriate of potash at the rate of 300 pounds per acre. These materials have been applied annually in accordance with the plan, except for the oat crop in 1911 when no fertilizers were applied. Also, through an error the nitrogen was omitted from plots 63 and 64 in 1914.

Each year a record is made of the kinds of weeds and grasses found on the uncultivated part of the plots, and the number of such plants recorded or the area which they cover measured. It is thus possible to say whether certain species are disappearing and others taking their places under the different treatments. In the late summer the weeds and grass are cut down on these plots but nothing is removed.

It is not the intention to give in this paper a detailed statement of the crop yields on the cultivated section nor to give a complete record of the vegetation on the uncultivated section (the latter is reserved for a later publication) but rather to compare the nitrogen and carbon content of the soils from the two sections, and, taking into consideration the nitrogen removed by the crops on the cultivated sections, determine whether the net result with reference to this element has been loss or gain. No account has been taken of the carbon in the crop so that a comparison so far as this element is con-

CULTIVATED HALF

57 Phos. Acid Potash	58 Phos. Acid Potash	59 Nitrogen Potash	60 Nitrogen Potash	61 Nitrogen Phos. Acid	62 Nitrogen Phos. Acid	63 Nitrogen Phos. Acid Potash	64 Nitrogen Phos. Acid Potash
57	58	59	60	61	62	63	64

UNCULTIVATED HALF

cerned is with reference to the amount in the soil of the two sections, cultivated and uncultivated.

Unfortunately no samples were collected from these plots when the work was begun.

Table 1 gives the results of nitrogen and carbon determinations made on samples taken from the cultivated and uncultivated sections of plots 57 to 64 in 1913 and also in 1916. In taking the samples 6 borings, to a depth of 6½ inches, were made with a tube cutting a core about 2 inches in diameter, and these cores were made into one sample, dried, passed through a 3-mm. sieve to remove coarse material and finally ground to pass a 0.5 mm. sieve.

Nitrogen determinations were made by the Kjeldahl-Gunning method, five or six determinations being made on each sample.

A study of this table brings out the fact that without exception both nitrogen and carbon are now higher in the uncultivated sections than in the cultivated. The average difference in nitrogen content between the two sections for the

nitrogen-treated plots, was 0.02 per cent in 1913, and 0.0234 per cent in 1916, the former being equivalent to 400 pounds of nitrogen per acre 6½ inches (2,000-000 pounds of soil) and the latter equivalent to 468 pounds. The average difference between the cultivated and uncultivated sections of plots 57 and 58, the plots that receive no nitrogen, was in 1913, 0.0123 per cent, which makes a difference in favor of the uncultivated section of 226 pounds of nitrogen per acre, whereas the difference in 1916 was 0.0258 per cent or 516 pounds per acre.

TABLE 1
Nitrogen and carbon in soil, 1913 and 1916

	TOTAL NITROGEN				TOTAL CARBON			
	1913		1916		1913		1916	
	Cultivated	Unculti-vated	Cultivated	Unculti-vated	Cultivated	Unculti-vated	Cultivated	Unculti-vated
57	0.0913	0.1054	0.0749	0.1003	1.028	1.320	1.003	1.275
58	0.0787	0.0891	0.0715	0.0977	0.995	1.130	0.995	1.269
Average.....	0.0850	0.0973	0.0732	0.0990	1.012	1.225	0.999	1.272
59	0.0809	0.0949	0.0802	0.0985	1.047	1.265	1.048	1.300
60	0.0913	0.1034	0.0824	0.1063	1.142	1.279	1.086	1.311
61	0.0795	0.1041	0.0841	0.0981	0.972	1.245	0.960	1.231
62	0.0874	0.1106	0.0773	0.1134	1.022	1.459	0.999	1.533
63	0.0806	0.1053	0.0802	0.1067	1.028	1.281	1.017	1.440
64	0.0876	0.1108	0.0856	0.1072	1.042	1.364	1.106	1.228
Average.....	0.0846	0.1049	0.0816	0.1050	1.042	1.316	1.036	1.341

This accords well with the view that liberal applications of the minerals favor the appearance of volunteer leguminous plants on abandoned land and also the fixation of atmospheric nitrogen by non-symbiotic bacteria, and no doubt accounts for the fact that on the uncultivated sections of these plots the nitrogen supply is fairly well maintained. On the other hand, the cultivated sections are gradually being depleted of their supply of nitrogen, the volunteer leguminous plants not being permitted to come in to restore the losses.

Taking an average of all the nitrogen-treated plots, the cultivated portions show slightly less nitrogen in 1916 than in 1913, in spite of the fact that they have received fair annual applications of nitrogenous fertilizers. For the uncultivated sections the average is essentially the same in 1916 as in 1913, that is, the gain during the three years has not been sufficient to make an appreciable difference in the analyses. It is evident that the total carbon is being gradually decreased on the cultivated sections. This is shown by an average percentage of 1.042 in 1913 and 1.036 in 1916. It is further emphasized,

however, by the fact that determinations made on samples collected in 1909 from nearby plots, where the soil is the same as the soil of the plots under consideration, show 1.21 per cent of total carbon (6). From this it would appear that the cultivated plots have lost nearly 0.2 per cent of carbon during the period 1909 to 1916. On the other hand, the carbon is undoubtedly

TABLE 2
Yields of nitrogen in a five-year rotation, 1908 to 1916 inclusive
(Calculated on the acre basis)

	CORN 1908	OATS 1909	WHEAT 1910	OATS 1911	TIMOTHY 1912	TOTAL NITROGEN FOR FIRST FIVE YEARS	CORN 1913	OATS 1914	WHEAT 1915	TIMOTHY 1916	TOTAL NITROGEN 1913-1916 INCLUSIVE	TOTAL NITROGEN 1908-1916 INCLUSIVE
57	52.76	19.44	25.48	17.80	5.64	121.12	30.44	14.76	18.60	18.55	82.35	203.47
58	45.60	17.28	28.12	18.16	2.16	111.32	28.64	12.14	19.20	17.49	77.47	188.79
Average.....						116.22					79.91	196.13
59	74.40	28.48	47.80	21.20	5.28	177.16	50.44	23.04	30.00	23.71	127.19	304.35
60	76.40	33.52	55.88	33.16	6.76	205.72	53.68	36.42	37.44	24.83	152.37	358.09
Average.....						191.44					139.78	331.22
Average increase over check.....						75.22					59.87	135.09
61	90.28	31.20	52.12	34.52	7.12	215.24	36.88	23.42	38.52	28.42	127.24	342.48
62	41.12*	33.76	46.72	30.00	6.44	158.04	51.40	25.92	32.38	27.14	136.84	294.88
Average.....						186.64					132.04	318.68
Average increase over check.....						70.42					52.13	122.55
63	77.04	30.24	51.92	33.84	7.00	200.04	58.40	22.48	29.72	29.01	139.61	339.65
64	74.36	28.88	53.44	36.12	9.84	202.64	54.64	21.22	35.42	26.26	137.54	340.18
Average.....						201.34					138.58	339.92
Average increase over check.....						85.12					58.67	143.79
General												
Average.....	72.27	31.01	51.31	31.47	7.07	193.14	50.91	25.41	33.91	24.90	136.80	329.94
Average increase over check.....						76.92					56.89	133.81

* Very low yield of stalks for some reason.

increasing in the uncultivated plots, as is shown by an average of 1.316 per cent in 1913 and an average of 1.341 per cent in 1916, and by the further fact, already mentioned, that the samples collected in 1909 from nearby plots show only 1.21 per cent of carbon. Thus it would appear that during the period 1909 to 1916 the carbon content of the uncultivated soils has increased by about 0.13 per cent, which would be equivalent to slightly more than 1½ tons

of carbon per acre 6½ inches. This is as one might expect, since each year there has been a fair growth of weeds and grasses on these plots and nothing has been removed.

Table 2 shows the amount of nitrogen removed by the crops on the cultivated portion of the plots for the years 1908 to 1916 inclusive, and the average amount for each crop on the eight plots. It will be observed that the corn crop of 1908 yielded the largest amount of nitrogen. This is followed by the corn crop of 1913 and the wheat crop of 1910, both of which furnished approximately 20 pounds less of nitrogen than the corn crop of 1908. The lowest yield was with the timothy of 1912. This is accounted for in part at least by the small crop of that year. With only two exceptions there was less nitrogen recovered from plots 57 and 58—the two that receive no nitrogen—than from those that receive an annual application of nitrogen. The average yield from plots 57 and 58 may be taken as the check, and if this is deducted from the average yields from the plots that receive nitrogenous fertilizers, the remainder may be taken as the amount of the applied nitrogen that was recovered in the crop. The nitrogen-treated plots have received the dried fish at the rate of 600 pounds per acre, but on account of the omission in 1911 the total for the five years was 2400 pounds instead of 3000 pounds. This, on the basis of 8 per cent nitrogen for the fish, would mean a total of 192 pounds of nitrogen per acre for the first five years and also 192 pounds for the last four years, excepting plots 63 and 64 which, on account of the omission in 1914, received only 144 pounds, thus making the average for the four years 176 pounds instead of 192 pounds. This gives a total for the nine years of 368 pounds.

These figures divided into the increase over the check for the respective periods, with the proper pointing off, give the percentages of nitrogen recovered in the crop.

The results calculated for the different treatments are as follows:

PLOTS	FIRST PERIOD, FIVE YEARS	SECOND PERIOD, FOUR YEARS
59.60	39.18	31.18
61.62	36.67	27.15
63.64	44.33	40.74
Average.....	40.06	33.02

Stated in another way the average increase over the check is, for the nine years, 133.81 pounds of nitrogen per acre (bottom figure in last column of table 2) and the total amount of nitrogen applied per acre during that time was 368 pounds. From these figures it is found that there was an average annual recovery for the nine years of 36.36 per cent of the applied nitrogen; that is, a recovery of only a little more than one-third. The highest average recovery for 9 years is from plots 63 and 64 which receive the complete fer-

tizer, and the next highest from lots 59 and 60 which receive nitrogen and potash.

The average recovery for the first period is a little higher than for the second period. These figures correspond quite closely with recoveries obtained from dried blood in cylinder experiments previously reported (7) and also for the most part with recoveries from fish scrap used in field experiments (6) where the rotation was the same as the one here considered. Slightly higher recoveries were obtained where fish was used as a source of nitrogen in pot experiments (8). The average total recovery from plots 59 to 64 for the nine years is 329.94 pounds (acre basis) while the total amount applied to each plot during the same period was 368 pounds if 8 per cent is taken as the analysis of the fish. During this period the crops on plots 57 and 58, without commercial nitrogen, were able to take from the soil an average of nearly 200 pounds of nitrogen per acre.

From the results it seems quite clear that nitrogen is the limiting factor in crop production on this soil, otherwise plot 60, where no potash has been applied, and plot 61, where no phosphoric acid has been applied, would hardly have given yields of nitrogen in excess of plots 63 and 64 where all three constituents were used. It is true that the averages for 59 and 60 and for 61 and 62 are not quite as high as the average for 63 and 64, but the difference is so small that one could hardly say that the yield was limited by a lack of potash in the one case or of phosphoric acid in the other.

That nearly two-thirds of the applied nitrogen should be unaccounted for is discouraging and does not plead very strongly for the use of commercial nitrogenous materials of an organic nature, if those of mineral origin are available.

SUMMARY

It is here pointed out that the average nitrogen content of land which was allowed to run wild from 1908 to 1916, and which during that period received annual applications of dried fish amounting to 600 pounds per acre, was essentially the same in 1916 as in 1913. (No samples were collected previous to 1913.)

The carbon content of this same land was increased slightly during the period 1913 to 1916.

The average nitrogen content of adjoining cultivated plots, under a 5-year rotation, was 0.02 per cent less in 1913, and 0.023 per cent less in 1916, than the nitrogen content of the corresponding plots that were allowed to run wild.

The average nitrogen content of the cultivated plots was slightly less in 1916 than in 1913.

The average carbon content of the cultivated plots was approximately 0.27 per cent less in 1913 and 0.30 per cent less in 1916 than the average carbon content of the corresponding plots allowed to run wild.

The average carbon content of the cultivated soils was slightly less in 1916 than 1913.

The two cultivated plots which received no nitrogen (the check plots) yielded, in the crops from these plots, an average total of 196.13 pounds of nitrogen for the nine years.

The six nitrogen-treated plots yielded, during the same period, an average total of 329.94 pounds of nitrogen in the crops from these plots.

There was recovered from the six nitrogen-treated plots for the nine-year period, an average of 36.36 per cent of the nitrogen that was applied.

The percentage of nitrogen and carbon in the cultivated soils is decreasing, even where dried fish is applied at the rate of 600 pounds per acre annually.

The percentage of nitrogen in the soils allowed to run wild, and which have received annual applications of nitrogen, appears to run about constant, but the percentage of carbon is increasing slightly.

Much volunteer white clover is appearing on the two plots which receive minerals but no nitrogen.

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THE DETERMINATION OF SOIL PHOSPHORUS¹

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INTRODUCTION

Robinson (10) has recently reported a critical comparison of three methods for the determination of soil phosphorus, namely: fusion with alkali carbonates, the Washington method, in which the soil is evaporated to dryness with hydrofluoric acid previous to ignition, and the Fischer method, which is characterized by an ignition between two evaporation with aqua regia. He concluded that all three of these give accurate results, provided that the first two be modified so as to get rid of the organic matter without an accompanying loss of phosphorus. He implies that the failure to recover the full amount of phosphorus from soils by the Washington method is due to the volatilization of a part of it during ignition, which he states may be avoided by previously evaporating the samples with nitric acid, in order to dissolve "enough bases to hold the phosphoric acid during the ignition," and then carrying out the latter operation "in such a manner that there is no glowing." Using a soil containing 0.04 per cent of phosphoric acid and 0.29 per cent of organic matter he added a known amount of phosphoric acid and 0.5 gm. of sugar, hastily ignited the mixture to glowing and found "an apparent loss of from 30 to 11 per cent of the phosphorus present."

In the fusion method, preceded, in the case of soils high in organic matter, by evaporation with magnesium nitrate solution or with nitric acid and subsequent ignition, he mentions (10, p. 149) that there is "one inherent error as previously pointed out, i.e., the solubility of small amounts of ammonium phosphomolybdate in aqueous solutions of sodium nitrate" and that this difficulty can be overcome by precipitating the sesquioxides, which carry down with them the phosphoric acid, redissolving these, and again precipitating the phosphorus as ammonium phosphomolybdate.

The work of the author reported in this paper does not confirm the statement of Robinson (10, p. 148, foot-note 5) regarding the liability of serious loss of ammonium phosphomolybdate in the course of the ordinary deter-

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mination by fusion with alkali carbonate. Cain and Hostetter² (2, p. 251) in reporting the solubility of the yellow ammonium phosphomolybdate in the presence of vanadium salts did not include sodium nitrate in the list of reagents tested. They found ammonium phosphomolybdate, even in the presence of vanadium salts, to be far less soluble in solutions of nitrates than in neutral ammonium molybdate. Gortner and Shaw (4) have shown that vanadium, in the amounts ordinarily present in the soil, does not interfere with the determination of the soil phosphorus.

In the Fischer (3) method the author finds there is liable to be a large amount of phosphoric acid left in the insoluble residue, thus making the method appear as unsuitable for the determination of total phosphoric acid as the methods by extraction with strong nitric or hydrochloric acid.

Applying the ordinary Washington (11, p. 162) method to soils the author finds that it is liable to give results much too low, but that this is not due to volatilization of phosphoric acid during ignition and is not lessened by previous evaporation with either magnesium nitrate solution or nitric acid, while the elimination of the organic matter by ignition without glowing, as proposed by Robinson (10, p. 149) is not feasible, especially in the case of soils high in organic matter.

EXPERIMENTAL

Soils used

Four samples, two of mineral soil and two of peat (table 1), were used in this study. Both of the first two, A, a Carrington silt loam and B, a Fargo silt loam, from Rice County, Minnesota, were rich in organic matter, while A contained a much larger proportion of coarse particles and accordingly was apt to carry a larger proportion of its phosphoric acid as apatite inclusions in quartz grains. Of the two peat soils, C was low in lime, while the other, D, carried a moderate amount. Both contained about 90 per cent of organic matter.

All the samples were ground in an agate mortar so as to pass a silk bolting cloth of 0.25 mm. mesh. Blank determinations on the reagents, made parallel with those on the soils, were exposed to exactly the same chances of contamination from outside sources as the latter. In all cases the amounts found in the blanks have been deducted from the percentages reported.

² Robinson suggests that the reason Fischer found slightly higher quantities of phosphoric acid by his method than by the fusion method is "probably due to the difficulty of precipitating small amounts of ammonium phosphomolybdate in the presence of much NaNO_3 in the fusion method, since Cain and Hostetter (Jour. Soc. Chem. Indus., 4 (1912), 250) have shown that aqueous solutions of NaNO_3 have a strong solvent effect on the phosphomolybdate when vanadium is present." The reference proved to be other than the one cited, doubtless Jour. Indus. Engin. Chem., 4 (1912), 250, but, in this article, Cain and Hostetter do not report data to corroborate the above statement.

TABLE 1
Composition of soils used

SOIL	VOLATILE MATTER	NON-VOLATILE MATTER	CaO	NITROGEN	ORGANIC CARBON
	per cent	per cent	per cent	per cent	per cent
A.....	12.11	87.89		0.384	5.38
B.....	21.10	78.90		0.836	9.95
C.....	90.59	9.41	0.63	2.00	48.95
D.....	89.06	10.94	1.86	2.47	48.47

Fusion method

Soils A and B were analyzed by fusion with sodium carbonate, after ignition alone as well as after previous evaporation with magnesium nitrate. In the case of the former treatment the phosphoric acid was determined both

TABLE 2
Phosphoric acid as found by the different modifications of the fusion method

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN SEPARATED SILICA	TOTAL AMOUNT RECOVERED
<i>1. Fusion without evaporation with magnesium nitrate: Sesquioxides not precipitated</i>					
A.....	1	0.236	0	0.013	0.249
	2	0.236	0	0.006	0.242
	Average.....	0.236	0	0.009	0.245
B.....	1	0.319	0	0.019	0.338
	2	0.325	0	0.006	0.331
	Average.....	0.322	0	0.012	0.334
<i>2. Fusion without evaporation with magnesium nitrate: Sesquioxides precipitated</i>					
A.....	1	0.242	0	0.000	0.242
	2	0.248	0	0.000	0.248
	Average.....	0.245	0	0.000	0.245
B.....	1	0.325	0	0.012	0.337
	2	0.331	0	0.006	0.337
	Average.....	0.328	0	0.009	0.337
<i>3. Fusion after evaporation with magnesium nitrate: Sesquioxides not precipitated</i>					
A.....	1	0.229	0	0.013	0.242
	2	0.236	0	0.006	0.242
	Average.....	0.233	0	0.009	0.242
B.....	1	0.331	0	0.006	0.337
	2	0.338	0	0.000	0.338
	Average.....	0.334	0	0.003	0.337

with and without the precipitation of the sesquioxides. In the case of each analysis the separated silica was evaporated with hydrofluoric acid and the phosphoric acid so obtained determined separately. In all the analyses the filtrate from the yellow precipitate was rendered alkaline with ammonia, the precipitate filtered off, redissolved and the phosphorus determined in this. The data are reported in table 2. The evaporation with magnesium nitrate was accomplished by dissolving 1 gm. of the salt in a few cubic centimeters of water and pouring this over the soil, after which it was evaporated to dryness and carefully ignited.

It is to be seen that only a very small amount of phosphoric acid was found in the separated silica and no advantage was gained from treatment with magnesium nitrate previous to ignition, or by precipitation of the sesquioxides to eliminate the excess of sodium salts.

Fischer method

This, which is a modification of the methods by acid extraction and designed to recover practically the whole of the phosphoric acid in soils, is characterized by two treatments with aqua regia. It is briefly as follows:

Five to ten gm. of dry soil and 50 cc. of aqua regia are digested on the steam bath in a covered silica dish until effervescence, if any, ceases, after which the watch glass is removed and the contents are evaporated to dryness and ignited until the residue turns brown. The contents are again evaporated to dryness with 50 cc. of aqua regia and the residue treated with concentrated nitric acid and evaporated to dryness to the disappearance of the hydrochloric acid. The residue is finally taken up with dilute nitric acid, filtered and the phosphorus determined in the filtrate.

To test the completeness of extraction by his method, Fischer (3, p. 546) used 4 soils varying in phosphoric acid from 0.14 to 0.47 per cent and found in the residues, which were fused with sodium carbonate, only from 0.004 to 0.009 per cent, a negligible quantity. He makes no mention of heating the residue after evaporation with nitric acid in order to dehydrate the silica. Without such a heating filtration is very difficult. The author, in testing this method, has, like Robinson (10, p. 148), heated the mass until it was brown and then extracted it with nitric acid. It is much better to heat the dish by carefully turning it in an open flame than by using a hot plate or sand bath, thereby avoiding the formation of compounds of iron and aluminum insoluble in nitric acid.

Soils A and B were analyzed by this method, the residue being fused with sodium carbonate. In the case of B the amount of phosphoric acid remaining in the residue in four determinations fell within the limits found by Fischer and in three others it varied from 0.013 to 0.019 per cent, while in each of six determinations with A the amount in the residue lay between the limits of 0.019 and 0.051 per cent, or from 8 to 20 per cent of the total amount present, the latter being far from a negligible quantity.

Suspecting that the large amount of phosphoric acid in the residues might be due to an over-heating after the final evaporation with nitric acid, the product thus obtained was heated in an air bath at 150°C. for 2 hours, and also at 110°C. for 1 hour. The data (table 3, parts 1 and 2) show that nothing

TABLE 3
Phosphoric acid as found by Fischer's method

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
<i>1. Dish heated over open flame</i>					
A.....	1	per cent 0.211	per cent 0	per cent 0.025	per cent 0.236
	2	0.217	0	0.019	0.236
	3	0.223	0	0.025	0.248
	3	0.185	0	0.051	0.236
	5	0.217	0	0.025	0.242
	6	0.213	0	0.038	0.251
	Average.....	0.211	0	0.038	0.241
B.....	1	0.325	0	0.000	0.325
	2	0.331	0	0.000	0.331
	3	0.331	0	0.000	0.331
	4	0.319	0	0.000	0.325
	5	0.319	0	0.013	0.332
	6	0.319	0	0.019	0.338
	7	0.306	0	0.019	0.325
Average.....		0.321	0	0.008	0.329
<i>2. Dish dried in oven for 2 hours at 150°C.</i>					
A.....	1	0.204	0	0.045	0.249
	2	0.217	0	0.019	0.236
	Average.....	0.211	0	0.032	0.243
B.....	1	0.312	0	0.019	0.331
	2	0.306	0	0.032	0.338
	Average.....	0.309	0	0.025	0.334
<i>3. Dish dried in oven for 1 hour at 110°C.</i>					
A.....	1	0.217	0	0.032	0.249
B.....	1	0.306	0	0.026	0.332

is gained by heating at a lower temperature, while this has the disadvantage of consuming more time. From the above it appears that while with some soils the method may give, at least under certain conditions, satisfactory results, with others it is likely to give results much too low.

Washington method

The soils A and B were analyzed by this method (11, p. 162), modified to the extent that after the second evaporation with nitric acid the residue, instead of being heated until it became brown, was ignited at a dull red heat. This was necessary in order to get rid of the organic matter. The method when applied to rocks, for which it was designed, does not need any such modification. The residue from the extraction with hydrofluoric and nitric acids was fused, as Hillebrand (7, p. 144) suggests, with sodium carbonate, the melt

TABLE 4
Phosphoric acid as found by Washington's method

DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
<i>Soil A</i>				
1	per cent 0.197	per cent 0	per cent 0.061	per cent 0.258
2	per cent 0.191	per cent 0	per cent 0.061	per cent 0.252
3	per cent 0.191	per cent 0	per cent 0.064	per cent 0.255
4	per cent 0.197	per cent 0	per cent 0.051	per cent 0.246
5	per cent 0.178	per cent 0	per cent 0.064	per cent 0.242
6	per cent 0.191	per cent 0	per cent 0.064	per cent 0.255
7	per cent 0.185	per cent 0	per cent 0.057	per cent 0.242
8	per cent 0.191	per cent 0	per cent 0.045	per cent 0.236
9	per cent 0.191	per cent 0	per cent 0.057	per cent 0.248
Average.....!	per cent 0.190	per cent 0	per cent 0.058	per cent 0.248
<i>Soil B</i>				
1	per cent 0.286	per cent 0	per cent 0.045	per cent 0.331
2	per cent 0.331	per cent 0	per cent 0.000	per cent 0.331
3	per cent 0.306	per cent 0	per cent 0.032	per cent 0.338
4	per cent 0.306	per cent 0	per cent 0.025	per cent 0.335
5	per cent 0.306	per cent 0	per cent 0.045	per cent 0.351
6	per cent 0.293	per cent 0	per cent 0.038	per cent 0.331
Average.....	per cent 0.303	per cent 0	per cent 0.031	per cent 0.336

dissolved and the resulting solution evaporated to dryness twice with nitric acid, the residue taken up with nitric acid and the phosphorus determined in the solution.

The data in table 4 shows that an appreciable amount of phosphoric acid remained in the residue in 14 out of 15 determinations, varying with A from 2 to 25 per cent of the total amount and with B amounting to about 10 percent.

The amount from the hydrofluoric acid extract and the residue is, within the limits of experimental error, equal to that found by the fusion (table 2) and modified Fischer methods (table 3). Accordingly, the low percentages

of phosphorus that have been found by this method are to be attributed not to volatilization during ignition but to the incompleteness of the extraction by nitric acid.

Both soils were also analyzed by Robinson's modification of the Washington method, in which the soil is evaporated with nitric acid previous to ignition, and any glowing in the latter operation is avoided. From 50 to 65 per cent was thus recovered (column 4 in table 5).

The residues were fused with sodium carbonate and otherwise treated as described in the preceding paragraph and about 20 per cent of the total phosphoric acid in the case of sample A and from 17 to 38 per cent in that of B was obtained (column 6, table 5), leaving still a deficit of 12 to 20 per cent of the total. Since the filtrate from the ammonium phosphomolybdate was strongly colored by organic matter left behind by the low temperature of ignition, it was thought this might still contain phosphorus. This was digested with aqua regia on the sand bath until the organic matter was en-

TABLE 5
Phosphoric acid as found by Robinson's modification of Washington's method

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT		IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
		First ignition	Second ignition			
A...	1	0.185	0.147	0.051	0.057	0.255
	2	0.172	0.147	0.051	0.051	0.249
	Average....	0.178	0.147	0.051	0.054	0.252
B...	1	0.185	0.159	0.045	0.127	0.331
	2	0.299	0.217	0.057	0.057	0.331
	Average....	0.242	0.188	0.051	0.092	0.331

tirely destroyed, after which it was made alkaline with ammonia, the sesquioxides filtered off, dissolved in nitric acid and the phosphorus reprecipitated with ammonium molybdate and determined as usual. The additional quantity thus removed accounts for the total amount (column 5, table 5). The filtrate from the yellow precipitate was tested for phosphorus for every method tried and the present instance was the only one in which any was found.

Both the extract of the ignition product with nitric acid and the filtrate from the first precipitation of ammonium phosphomolybdate were colored, more strongly so with B than with A, while the filtrate from the second precipitation of ammonium phosphomolybdate, that obtained by dissolving the precipitated sesquioxides, was colorless. The first of the two yellow precipitates was dark-colored and this when dissolved in ammonia previous to the addition of magnesium mixture yielded a solution which was dark-colored, resembling the ammonia extract from a soil low in humus (*matière noire* of

Grandeau). The magnesium ammonium phosphate precipitated from this solution was gray as though containing some of the so-called magnesium humate. To decide whether magnesium other than that in the magnesium ammonium phosphate was contained in this precipitate the latter, after being filtered, washed, dried, ignited and weighed, was dissolved in nitric acid and reprecipitated with magnesium mixture. The phosphoric acid calculated from the weight of the resulting magnesium pyrophosphate was much less than that first obtained (columns 3 and 4, table 5).

The phosphoric acid was also determined on the samples A and B by this modification of the Washington method with the exception that after the first extraction with nitric acid they were ignited at a higher temperature, a *very dull red*, and then treated as Robinson directs, and as described above. The data are reported in table 6.

TABLE 6
Phosphoric acid as found by Robinson's modification of Washington's method; Samples ignited to a very dull red

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT		IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
		After first ignition	After second ignition			
A . .	1	0.229	0.185	0	0.064	0.249
	2	0.212	0.172	0	0.064	0.236
	Average....	0.220	0.179	0	0.064	0.243
B . .	1	0.299	0.261	0	0.083	0.344
	2	0.299	0.274	0		
	Average....	0.299	0.267	0		

In no case was the organic matter entirely oxidized by heating to this temperature, although its elimination was undoubtedly more complete in the portions of the sample which were in contact with the dish. In such parts the heat may have been sufficient not only to oxidize the organic matter, but also to produce undesirable changes, while in adjacent parts the oxidation would still be incomplete. The solutions obtained by extracting the ignition product with nitric acid were all colored, although those from A were only slightly so. The filtrates from the ammonium phosphomolybdate were slightly colored but contained no phosphoric acid.

The data in tables 5 and 6 indicate that with soils high in organic matter the magnesium pyrophosphate obtained by Robinson's modification of Washington's method contains a considerable amount of magnesium oxide. Furthermore, a large part of the phosphoric acid may not be precipitated by the ammonium molybdate, probably due to its presence in organic compounds which in turn are due to incomplete oxidation at the low temperature of ignition necessary to avoid all glowing. Further, Robinson's modification of

this method does not appear in any way to affect the amount of phosphorus in the residue from the hydrofluoric acid extraction. It is of interest to note that in the case where the samples were ignited to a very dull red the errors of the method are nearly compensating (columns 3 and 7, table 4). It would also appear that Robinson's modification is far less reliable for soils than Washington's original method modified simply to the extent of igniting the soil to dull redness after the third evaporation with nitric acid.

As it may be suspected that the ignition at a temperature below glowing was faulty, a brief description of this operation as practised by the author

TABLE 7

Phosphoric acid in peat soils as found after ignition both with and without previous evaporation with magnesium nitrate solution

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
<i>1. Without previous treatment</i>					
C..... {	1	per cent 0.242	per cent 0	per cent 0.013	per cent 0.255
	2	0.242	0	0.013	0.255
	Average.....	0.242	0	0.013	0.255
D..... {	1	0.289	0	0.006	0.295
	2	0.283	0	0.013	0.296
	Average.....	0.286	0	0.009	0.295
<i>2. Evaporated with magnesium nitrate solution</i>					
C..... {	1	0.242	0	0.013	0.255
	2	0.229	0	0.013	0.242
	Average.....	0.235	0	0.013	0.248
D..... {	1	0.293	0	0.006	0.299
	2	0.280	0	0.019	0.299
	3	0.280	0	0.000	0.299
	Average.....	0.291	0	0.008	0.299

appears desirable. The ignition was carried out in a somewhat darkened part of the room and, as a further precaution, the dish was shaded with a large piece of cardboard. Held by means of a pair of platinum-tipped tongs the dish was turned round above a Bunsen flame burning at approximately half its capacity so that every part was exposed to the heat. As soon as any portion showed the faintest indication of reddening the dish was turned so as to allow the heat to strike a cooler part.

From the above there appears no good reason to attribute any incomplete recovery of phosphorus to loss by volatilization during ignition. Additional evidence is presented in table 7 showing the amount of phosphoric acid in

two peat soils recovered both by the ordinary ignition to redness without any previous treatment and by a similar ignition after evaporation with magnesium nitrate solution. As both samples carried 90 per cent of organic matter any loss of phosphorus due to lack of bases should have been expected in C which carried so little lime that unless previously treated with ground limestone or some other form of agricultural lime it has regularly proved sterile to all ordinary crops, except redtop (*Agrostis alba*). In C as well as in D no increase in the amount of phosphoric acid recovered results from the treatment with magnesium nitrate.

As further evidence of this same fact sample A was mixed with 0.5 gm. of sugar and ignited, not *hastily* but carefully, as in the ordinary determination, and the phosphoric acid determined by the proposed modification (see below) of Washington's method. The data are shown in table 8. There appears no justification for a hasty ignition especially when there is so great an opportunity for mechanical loss along with the escape of the water of

TABLE 8
Phosphoric acid as found in Sample A after ignition with sugar

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT
A..... {	1	per cent 0.248	per cent 0	per cent 0.006	per cent 0.254
	2	per cent 0.248	per cent 0	per cent 0.006	per cent 0.254
	Average.....	per cent 0.248	per cent 0	per cent 0.006	per cent 0.254

decomposition from the sugar. In case a solution containing phosphorus had been added the chance for loss in this manner would have been still greater.

Author's modification of Washington method

The author has found that Washington's method may be modified slightly so as to provide for the elimination of the organic matter of soils and the complete extraction of the phosphorus by means of hydrofluoric acid and nitric acid without making it less satisfactory for soils than it has proven for rocks. The only place where it would not be suitable would be where platinum ware is not available.

The dried soil is, as in the methods of Grandjeau (5, p. 267) and Hilgard (6, p. 20), ignited to dull redness, preferably in a muffle, and then treated according to Washington's directions up to the last evaporation with nitric acid. Then the residue in the platinum dish is heated in an air bath at 110°C. for 1 or 2 hours in order to dehydrate the silica, the greater length of time being preferable since a more complete dehydration is obtained and filtration is rendered easier. It may even be satisfactorily heated over a naked flame of the Bunsen burner, but this is not to be recommended, as there is a much

greater danger of overheating than by drying the sample in an oven. Filtration after dehydration of the silica by the latter method, however, is considerably slower than after dehydration above the direct flame. Using twelve samples dried in an air bath for 2 hours at 110°C. the time for filtration and washing was determined and found to be 2 hours.

TABLE 9
Phosphoric acid as found by the author's modification of Washington's method

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN FILTRATE FROM YELLOW PRECIPITATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
<i>1. Heated in an air bath 1 hour at 110° C.</i>					
A..... {	1	per cent 0.242	per cent 0	per cent 0	per cent 0.242
	2	0.248	0	0	0.248
	Average.....	0.245	0	0	0.245
B..... {	1	0.331	0	0	0.331
	2	0.331	0	0	0.331
	Average.....	0.331	0	0	0.331
<i>2. Heated 1 to 2 hours in an air bath at 150° C.</i>					
A..... {	1	0.204	0	0.045	0.249
	2	0.217	0	0.019	0.236
	Average.....	0.211	0	0.032	0.243
B..... {	1	0.312	0	0.019	0.331
	2	0.306	0	0.032	0.338
	Average.....	0.309	0	0.025	0.334
<i>3. Heated above an open flame</i>					
A..... {	1	0.255	0	0.000	0.255
	2	0.217	0	0.025	0.242
	Average.....	0.236	0	0.012	0.248
B..... {	1	0.338	0	0.000	0.338
	2	0.344	0	0.000	0.344
	Average.....	0.341	0	0.000	0.341

The formation of red oxide insoluble in nitric acid is evidence of overheating. In case such a red oxide is formed a few cubic centimeters of nitric acid and hydrofluoric acid are added to the platinum dish containing the sample, the contents evaporated to dryness and the evaporation repeated twice with nitric acid to decompose the fluorides, after which it is treated as a new sample.

The method as finally developed is briefly as follows:

One gram of dry soil is weighed out in a platinum dish of appropriate size and ignited in the muffle to a dull red, being maintained at this temperature a sufficient length of time to insure a complete oxidation of the organic matter. After cooling, 10 cc. of distilled water, 10 cc. of nitric acid and 5 cc. of hydrofluoric acid are added, the mixture well stirred, and the contents of the dish evaporated on the steam bath until approximately 5 cc. remain, when an additional 5 cc. of hydrofluoric acid are added, the mixture again well stirred, and the evaporation continued to complete dryness. Evaporation with small quantities of nitric acid is to be repeated two or three times. After the final evaporation the residue is dried in the air bath for 1 or 2 hours at 110°C. in order, as Washington (11, p. 163) states, "to render insoluble any silica which might otherwise come down with the phosphorus." When cool the residue is taken up with 3 cc. of concentrated nitric acid and 7 cc. of distilled water, boiled gently for a few minutes, and after cooling somewhat, is filtered and washed; the phosphorus in the filtrate is precipitated with ammonium molybdate,⁸ and finally weighed as magnesium pyrophosphate. With soils poor in phosphorus a larger sample of soil should be taken and the amounts of the reagents proportionally increased.

The cause of the phosphoric acid remaining in the residue when this (after evaporation of the nitric acid) has been heated too strongly appears to be due to a reaction with iron and aluminum salts to form difficultly-soluble phosphates. Grandjeau (5, p. 268) called attention to the fact that if the residue left after evaporation with nitric acid is heated to glowing, the oxides of iron and aluminum combine with small amounts of phosphoric acid and are only sparingly soluble in nitric acid. The author, using solutions containing known amounts of iron as ferrous ammonium sulfate, aluminum as aluminum sulfate and phosphoric acid as sodium phosphate, added hydrofluoric and nitric acids and continued the analysis according to Washington's method, including an ignition at dull redness previous to final treatment with nitric acid. This was done both with the solution alone in different amounts and with it added to soils A and B. As will be seen from table 10 the phosphoric acid was not completely recovered, but the deficit was fully accounted for by the amount obtained in the residue by fusion with sodium carbonate. The amount in the residue constituted in all cases a large part of the whole. There was no definite increase in this proportion due to the use of a larger amount of the solution of iron and aluminum.

Using the same soils and solutions, the determinations were made also by the author's modification of the Washington method, the results being given in the second part of the same table. All the phosphoric acid was recovered in the nitric acid extract, the residue retaining either no phosphoric acid or at most practically a negligible quantity.

Hilgard's method

The Hilgard method (6) for the determination of the acid-soluble phosphoric acid, which consists of the ignition of the soil and then digestion with concentrated nitric acid on the water bath for 2 days, gives as complete an

⁸ An acid solution of ammonium molybdate was used throughout this study.

TABLE 10

Phosphoric acid as found after the addition of known amounts of iron; alum num, and phosphorus

SOIL	DETERMINATION	CONTENT OF SOLUTION ADDED, EXPRESSED AS PER CENT OF SOIL			PHOSPHORIC ACID			TOTAL AMOUNT RECOVERED
		Fe ₂ O ₃	Al ₂ O ₃	P ₂ O ₅	In soil	In nitric acid	In residue	

1. By Washington's method

	Solution 1	2.0	5.0	1.020	per cent	0.944	0.057	1.001
	2	2.0	5.0	1.020		0.914	0.096	1.010
	Average.....	2.0	5.0	1.020		0.929	0.077	1.006
A....	1	2.0	5.0	1.020		1.178	0.070	1.248
	2	2.0	5.0	1.020		1.167	0.108	1.275
	Average.....	2.0	5.0	1.020	0.245	1.173	0.089	1.262
B....	1	2.0	5.0	1.020		1.275	0.070	1.345
	2	2.0	5.0	1.020		1.282	0.057	1.339
	Average.....	2.0	5.0	1.020	0.334	1.278	0.064	1.342
	Solution 1	5.0	10.0	1.020		0.860	0.153	1.013
A....	1	5.0	10.0	1.020		1.170	0.089	1.259
	2	5.0	10.0	1.020		1.190	0.096	1.286
	Average.....	5.0	10.0	1.020	0.245	1.180	0.092	1.272
B....	1	5.0	10.0	1.020		1.230	0.134	1.364
	2	5.0	10.0	1.020		1.230	0.121	1.351
	Average.....	5.0	10.0	1.020	0.334	1.230	0.127	1.357

2. By author's modification of Washington's method

	Solution 1	2.0	5.0	1.020		1.020	0.000	1.020
	2	2.0	5.0	1.020		1.026	0.000	1.026
	Average.....	2.0	5.0	1.020		1.023	0.000	1.023
A ...	1	2.0	5.0	1.020		1.262	0.006	1.268
	2	2.0	5.0	1.020		1.243	0.019	1.262
	Average.....	2.0	5.0	1.020	0.245	1.252	0.013	1.265
B....	1	2.0	5.0	1.020		1.345	0.000	1.345
	2	2.0	5.0	1.020		1.332	0.012	1.344
	Average.....	2.0	5.0	1.020	0.334	1.339	0.006	1.345

The author has found in using an artificial soil solution containing ferrous ammonium sulfate equivalent to 5 per cent Fe₂O₃, aluminum sulfate equal to 10 per cent Al₂O₃ and sodium phosphate equal to 0.255 per cent P₂O₅ (expressed as per cent of soil) that it required the addition of an amount of

extraction as the Washington method when the latter is used without taking the precaution of avoiding over-heating the residue from the nitric acid extraction (table 4). The completeness of the extraction by this method is of especial interest on account of the great number of determinations carried out in the laboratory of its famous author and the valuable conclusions based upon these. Were there an actual danger of loss of phosphoric acid by volatilization through ignition before digestion much of his data would doubtless have been far too low. However, as it has been shown that there is no reason to suspect loss by volatilization we have no reason to question the reliability of his data. Practically as much phosphorus was extracted from sample A by this method as by the Fischer method, but contrary to what would be expected, considerably less from sample B.

TABLE 11
Phosphoric acid as found by Hilgard's method

SOIL	DETERMINATION	IN NITRIC ACID EXTRACT	IN RESIDUE	TOTAL AMOUNT
		per cent	per cent	per cent
A.....	1	0.210	0.025	0.235
	2	0.210	0.032	0.242
	Average.....	0.210	0.029	0.239
B.....	1	0.293	0.032	0.325
	2	0.286	0.038	0.324
	Average.....	0.290	0.035	0.325

A summary of the results obtained by the different methods is shown in table 12.

Influence of titanium

The presence of titanium has been regarded as tending to retard the precipitation of ammonium phosphomolybdate (8, p. 594). Little work on the subject has been reported and the most of that done has been in connection with the determination of phosphorus in steel or in iron ore, where the percentage is relatively high when compared with the amount usually found in soil.

titanium sulfate equivalent to 2.5 per cent or more of TiO_2 to interfere seriously with the recovery of the phosphorus (table 13). A higher percentage seriously affected the accuracy of the determination.

Robinson (9, p. 12-13) in a study on 26 soils from 13 different types finds the percentage of TiO_2 to vary between 0.40 and 1.59 per cent with an average of 0.86 per cent for all, while the author (1, p. 422), in examining 36 samples of loess soil, found the upper and lower limits of TiO_2 present to be 1.30 and 0.96 per cent, respectively, with a general average of 1.09 per cent.

TABLE 12
Phosphoric acid as found by the different methods

METHOD	SOIL	NUMBER DETERMINATIONS AVERAGED	IN NITRIC ACID EXTRACT	IN FILTRATE FROM VARIOUS PHOSPHATE	IN RESIDUE	TOTAL AMOUNT RECOVERED
			per cent	per cent	per cent	per cent
Fusion unmodified.....	A	2	0.236	0.000	0.009	0.245
	B	2	0.322	0.000	0.012	0.334
Fusion sesquioxides precipitated.....	A	2	0.245	0.000	0.000	0.245
	B	2	0.328	0.000	0.009	0.337
Fusion Mg(NO ₃) ₂ modification.....	A	2	0.233	0.000	0.009	0.242
	B	2	0.334	0.000	0.003	0.337
Fischer.....	A	6	0.211	0.000	0.030	0.241
	B	7	0.321	0.000	0.008	0.329
Washington.....	A	9	0.190	0.000	0.058	0.248
	B	6	0.303	0.000	0.031	0.336
Washington, Robinson's modification.....	A	2	0.147	0.051	0.054	0.252
	B	2	0.188	0.051	0.092	0.331
Washington, author's modification.....	A	2	0.245	0.000	0.000	0.245
	B	2	0.331	0.000	0.000	0.331
Hilgard.....	A	2	0.210		0.029	0.239
	B	2	0.290		0.035	0.325

From data reported in table 13 it is evident that the amount of titanium usually present in soil, or even the maximum amount found by Robinson, is too low to affect the determination of the phosphorus.

TABLE 13
Phosphoric acid as found in a standard solution after the addition of known amounts of titanium

SOLUTION	CONTENT OF SOLUTION ADDED EXPRESSED AS PER CENT OF SOIL				P ₂ O ₅ RECOVERED		
	Fe ₂ O ₃	Al ₂ O ₃	TiO ₂	PrO ₅	Set 1*	Set 2	Average
1	5.0	10.0	1.0	0.255	0.255	0.248	0.251
2	5.0	10.0	1.5	0.255	0.255	0.255	0.255
3	5.0	10.0	2.5	0.255	0.236	0.242	0.239
4	5.0	10.0	5.0	0.255	0.147	0.197	0.172
5	5.0	10.0	10.0	0.255	0.019	0.032	0.025

* Sets 1 and 2 were run on different days.

SUMMARY

1. Six methods were compared, viz., fusion with sodium carbonate, the Fischer and Hilgard methods involving extraction with strong acid, Washington's hydrofluoric acid method, a modification of the last recently proposed by Robinson and a modification developed by the author.
2. With the fusion method usually only a negligible quantity of phosphoric acid was recoverable from the separated silica. Neither evaporation with magnesium nitrate previous to ignition nor precipitation of the phosphoric acid with the sesquioxides of iron and aluminum in order to separate it from the excess of sodium salts was found advantageous.
3. The Fischer method recovered practically all the phosphoric acid in the peat soils, but in most cases with mineral soils a considerable amount was left in the insoluble residue. None was lost by volatilization or rendered unrecoverable by the formation of compounds of iron and aluminum insoluble in nitric acid. That left in the residue was the result of an incomplete extraction by the acids employed.
4. The Hilgard method for the determination of acid-soluble phosphoric acid extracted as much from one soil as did the Fischer method, but in the case of another considerably less.
5. Washington's method, when applied to soils and modified to the extent of igniting the residue to dull redness after the final evaporation with nitric acid, failed to recover the whole of the phosphoric acid present. The residues upon being fused with sodium carbonate yielded the missing amount, thus showing that the low percentages found by this method are not due to volatilization during ignition but to incompleteness of extraction by nitric acid.
6. Robinson's modification of Washington's method extracted only from 50 to 65 per cent of the total phosphoric acid, the remainder being found partly in the residue and partly in the filtrate from the yellow precipitate. With soils high in organic matter the magnesium pyrophosphate obtained by this modification of the method carried a considerable amount of magnesium oxide, which was derived from precipitated organic compounds. A temperature so low that no glowing was produced failed to oxidize the organic matter completely. Samples analyzed by this modification of Washington's method with the exception that, after the first evaporation with nitric acid, they were ignited to very dull redness, behaved similarly, although no phosphoric acid was found in the filtrate from the yellow precipitate.
7. Two peat soils analyzed both with and without evaporation with magnesium nitrate solution previous to the ignition, gave no increased amount of phosphoric acid from treatment with this reagent, showing that there was no loss of phosphoric acid by volatilization during ignition. A sample of mineral soil mixed with 0.5 gm. of sugar and carefully ignited showed no loss of phosphorus by volatilization.

8. The incomplete extraction of the phosphorus from soils by Washington's method was found to be due to overheating the residue, causing the formation of difficultly-soluble phosphates of iron and aluminum.

9. The Washington method may be modified slightly so as to provide for the complete oxidation of the organic matter and the subsequent complete extraction of the phosphoric acid without making it less satisfactory for soils than it has proven for rocks. The prominent features of this proposed modification of the method consist of an ignition at dull redness previous to the treatment with hydrofluoric and nitric acids and later taking up the phosphoric acid with nitric acid, and drying the residue in an air bath at 110°C.

10. The amount of titanium oxide found in soils is too low to interfere with the precipitation of the phosphorus.

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HYDROGEN-ION CONCENTRATION MEASUREMENTS OF SOILS OF TWO TYPES: CARIBOU LOAM AND WASHBURN LOAM

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INTRODUCTION

In work recorded in an earlier paper (7) a number of soils of different type and origin were examined to determine the applicability of measurements of hydrogen-ion concentration to the soil. It was found possible to determine the hydrogen-ion concentration of soil extracts or soil suspensions in water by different methods: (1) an electrometric and (2) a simple colorimetric method. The results obtained by the two methods agreed sufficiently well to show that either method yields approximately correct results. The different soil samples exhibited widely different hydrogen-ion concentrations: the most acid soil showed the hydrogen-ion exponent¹ 4.4; the most alkaline, 8.6. More recently Sharp and Hoagland (12) have measured a variety of soils by the electrometric method and found exponents between the limits 3.7 and 9.7.

Wherry (16) has measured the hydrogen-ion concentration of some soils by the colorimetric method in a chemical study of the rocks and soils supporting the growth of the walking fern. Such soils showed exponents 5.5 to 9.0: the mean being 7.0.

Conner (5) estimated the intensity of acidity of soils by measuring the hydrolysis of ethyl acetate at the ordinary temperature. This method is not rapid, and the results which it yields have not yet been compared with those obtained by the above methods. Truog (13) has attempted to estimate the acidity by measuring the avidity of the "active" soil acids for bases: it is not clear just what relation this avidity would have to the hydrogen-ion concentration.

It will be recalled that the intensity of acidity is measured by the hydrogen-ion concentration, and that it bears in general no simple or direct relation

¹ Following the general custom, we report the "exponent" instead of the concentration itself. If desired, the concentration may be calculated by raising the number "10" to the power denoted by the exponent and finding the reciprocal of the result. Since, however, in practically all of the work on the biological significance of the hydrogen-ion concentration the results are stated in terms of the exponent, such a calculation is unnecessary. It need only be borne in mind that the exponent is about 7 for neutrality; and that the more alkaline, the greater is the value of the exponent; and the more acid, the lower the exponent.

to the quantity of acid present. It has been shown many times² that the hydrogen-ion concentration possesses a greater significance in biochemical processes than the quantity or concentration of acid substance. It is therefore entirely possible that certain problems of soil fertility, especially those relating to the necessity or desirability of liming for any specific crop, which have not as yet been solved by means of determinations of "lime requirement," may be solved by measurements of hydrogen-ion concentration.

The expression "to neutralize acidity" is ambiguous. It may mean to add alkali until there remains no excess of hydrogen ions over hydroxylions (physico-chemical standpoint); or it may mean to add alkali until acidic hydrogen atoms are all replaced (purely chemical or stoichiometrical standpoint), in which latter case the resulting reaction may be neutral, acid, or alkaline, depending on the nature of the salt formed. Most methods for estimating lime requirement, such as that of Veitch (14), are based on neutralization in the first sense; the method proposed by Bouyoucos (2) is based on neutralization in the second sense.

There is a current opinion that the rational liming of soils must be a process of neutralization, in one or the other of the above senses. This opinion seems not in accordance with the fact that many crops do well on soils regarded as acid; some, such as blueberries, have been found to require acid soils (6) others, such as potatoes, become liable to disease if liming is practiced. It would appear evident that what constitutes scientific corrective treatment of soil acidity, or soil alkalinity, cannot be settled by convention: we must find out what reaction (hydrogen-ion exponent) we ought to impart to the soil for any system of cropping, before we attempt to formulate rules for the adjustment.

SELECTION OF SOILS FOR EXAMINATION

In the earlier paper, cited above, no attempt was made to correlate the results found for the soils with the nature of the soils or with their biological characteristics, the soils under examination being too diverse. The measurements reported in this paper, on the other hand, have been made on soil samples grouped, according to the Bureau of Soils, into two types: Caribou loam and Washburn loam (15). The samples were taken from fields representative of large areas and conformed to one or the other of the two types.

Of the several kinds of soil upon which Irish potatoes are grown in northern Maine, the greater part of the crop is harvested from the Caribou loam and to a less extent from the Washburn loam. The types are quite similar in texture and are derived from the same parent materials, or glacial till; but, owing to certain topographic differences which affect particularly the local drainage, striking differences are found between soils of these types.

² See especially the monograph of Michaelis (11) and a recent paper by Clark and Lubs (4) for instances and literature.

The Caribou loam is characterized by the yellowish-brown color of the surface soil and even brighter subsurface soil (indicative of good drainage); whereas the surface of the Washburn loam is dark gray to black in color, and is underlaid by a light gray to a dull yellow and gray mottled subsurface soil, the latter being very characteristic of poorly-drained soils. The average depth of the surface soil of both types is about eight inches, which is probably one or two inches below the average plow depth. The Caribou loam occurs on high, well-drained ridges and slopes; the Washburn occurs in low-lying, flat or depressed situations, in which seepage or poor drainage naturally prevails. Such low situations favor the accumulation of muck and peat with which the Washburn loam is always covered until the land is cleared for cultivation. Similar mucky conditions do not occur on the virgin Caribou soils. The native vegetation of the Caribou loam is a hardwood growth of maple, yellow birch, white pine, balsam fir, and the like. The native timber growth of the Washburn loam is chiefly white cedar, except where the original timber has been destroyed by fire, when it is often replaced by other soft woods, such as poplar, white birch and cottonwood.

At least two-thirds of the potatoes grown in northern Maine are produced on the Caribou loam, which is in general better suited to potato culture than the Washburn loam. In connection with certain potato-disease studies carried on cooperatively by the Maine Agricultural Experiment Station and the United States Department of Agriculture, Schreiner, Lapham, and Westover in 1915 found that potato scab, particularly that caused by *Spongospora subterranea*, where it occurs at all, is largely on the Washburn loam, the Caribou loam being comparatively free from disease; and this has been confirmed by Hurst in 1916. An instance of the comparative freedom of the Caribou loam from infection of *Spongospora subterranea* has already been reported by Melhus, Rosenbaum, and Schultz (10). Soil 2.0 (p. 237-239) is the Caribou loam while 1.0 is the Washburn, the other numbers (1.1, 1.2, 2.1) being intermediate phases of the two types.

EXPERIMENTAL PROCEDURE

The procedure for the colorimetric determinations is much the same as that used before. We have found that acid soils may show a somewhat higher acidity when the soil extracts are made with 1 cc. of water per gram of air-dry soil than with 2 cc., the quantity used previously. Good duplication is obtainable with this ratio of 1 to 1. The average difference obtained by varying the ratio from 1 to 1, to 2 to 2 was 0.14 in the value of the exponent for nine soils. This indicates that at optimum moisture content the acidity may be more intense, but to what extent cannot be calculated. Only results between the same ratios of water to soil should be compared. We have used in the present work 1 cc. of water per gram of air-dry soil.

Most of the soil extracts (suspensions from which the heavier particles have been thrown out by means of a centrifuge) show an increased hydrogen-ion concentration when examined colorimetrically, if carbon dioxide is passed into them. If air which has been freed from carbon dioxide is blown through the extracts, some show very little change, others a moderate decrease of acidity, but all we have tried maintain their acidity to methyl red far longer than water, or suspension of bicarbonates, made acid to methyl red with carbon dioxide.*

When soils are dried in air, carbon dioxide is lost, for the pressure of carbon dioxide in the soil air is considerably higher than in the atmosphere. It has seemed more expedient for the present, however, to study air-dried soils.

The procedure has therefore its theoretical limitations, and the acidities found may be somewhat low for the two reasons given above.

About thirty grams of air-dry soil, which had been passed through a 1-mm. sieve without grinding, were treated in a centrifuge tube of 100 cc. capacity with an equal number of cubic centimeters of distilled water. The tube was closed by the hand, and after thorough wetting of the soil was accomplished, the tube was violently shaken about fifty times. Not more than eight soils were thus treated at a time and the entire determination was carried out without delay. The tubes were centrifuged for ten to twenty minutes. The supernatant fluid was drawn off with a pipette provided with a rubber tube and a mouth-piece and distributed into test-tubes. To a small portion a drop of methyl red was added; the result showed whether methyl red could be used for the measurement, if not, other indicators were tried. To a 5-cc. portion of the fluid the proper indicator solution was added and admixed, and the color so developed was compared with the colors obtained on adding the same quantity of the same indicator solution to tubes containing 5 cc. of various "buffer solutions" of known hydrogen-ion exponents.

Most of the fluids were somewhat turbid, and the simple comparator of Hurwitz, Meyer and Ostenberg (8), as modified by Clark and Lubs (4), was used for the comparison.

The buffer mixtures were made from fifth-molecular solutions of acid potassium phthalate, acid potassium phosphate, and sodium hydrate, according to the proportions given by Clark and Lubs (3, 4). The hydrogen-ion exponents

* It should not hastily be assumed that hydrogen-ion concentrations due to carbon dioxide are of less physiological significance than those due to other acids. The hydrogen-ion concentration of the blood, which is so important a quantity that a special mechanism involving the regulation of lung activity has been elaborated for keeping it constant, is maintained constant by suitably adjusted changes in the carbon dioxide content in spite of variations resulting from the accession of various acid or alkaline substances. (See the monograph of Michaelis for discussion and references.)

When carbon dioxide is accompanied by carbonates in analytically significant quantities the hydrogen-ion concentration is far lower for moderate pressures than those reported here for the more acid class of soils. [A general discussion of the carbonate equilibrium has been given by Johnston (9).]

of the buffer mixtures were checked electrometrically. If the substances used are known to be pure this is unnecessary.

The following two indicators were used; methyl red, and bromcresol purple (dibrom-o-cresol-sulfone-phthalein). Both were used in water solution: methyl red, two drops of a 0.02 per cent solution of the sodium salt; and bromcresol purple, three drops of a 0.04 per cent solution of the sodium salt. Although a slight excess of alkali is necessary in order to dissolve the indicators (4), if the excess is very slight over the theoretical quantity (one equivalent) the resulting solution's appear to have, if anything, less buffer action than the alcoholic solutions formerly used. At any rate the two solutions used by us gave concordant values (about 5.8) of the hydrogen-ion exponent for the distilled water used.

In one or two cases it was observed that even with the comparator the color of the soil extract treated with brom-cresol purple could not be matched in value exactly with any of the buffer solutions containing this indicator. This is due to dichromatism. Clark and Lubs have pointed out the disturbing effect of dichromatism in turbid solutions of this indicator and have shown how to reduce it (4). We have viewed the solutions through thin layers (12 mm.) in the comparator, but have not used the screened source of light devised by them. For the present differentiation the error due to dichromatism was unimportant: in fact, the mere differentiation could have been effected, as it proved, by the use of methyl red alone.

EXPERIMENTAL RESULTS

The results for cultivated fields of soils of the Caribou loam type are given in table 1, and the corresponding results for the Washburn soils in table 2. The samples were taken from the first 8 inches. The soils are arranged in order of decreasing acidity. The indicator used for the Caribou soils was methyl red, for the Washburn soils the indicator was also methyl red except for the last five cases.

TABLE 1
Hydrogen-ion exponents of cultivated soils belonging to the Caribou loam type

Soil number.....	1	2	3	4	5	6	7	8	9	10	11	12	13
Exponent.....	4.85	5.0	5.05	5.1	5.15	5.2	5.2	5.25	5.3	5.3	5.3	5.35	5.45

TABLE 2
Hydrogen-ion exponents of cultivated soils of the Washburn loam type

Soil number.....	14	15	16	17	18	19	20	21	22	23	24
Exponent.....	5.4	5.5	5.7	5.7	5.8	5.8	5.8	6.25	6.3	6.4	6.65

A further interesting comparison is noted when these results are tabulated according to the occurrence of both types on the same field. This is done in table 3.

TABLE 3

Hydrogen-ion exponents of soils of the Caribou loam and Washburn loam, collected in the same field

FIELD	CARIBOU LOAM	WASHBURN LOAM	DIFFERENCE
A	5.00	5.80	0.80
B	5.15	5.50	0.35
C	5.30	5.70	0.40
D	5.35	5.70	0.35
E	5.10	6.40	1.30
F	5.20	5.40	0.20
G	5.25	5.80	0.55
H	5.20	6.30	1.10
I	5.45	6.65	1.20

When one considers that the two types in each of these fields received exactly the same treatment in fertilization and cultivation, it shows that the differences are not accidental but are characteristic of the soil type. It should further be emphasized that these Maine potato soils have never been limed.

DISCUSSION OF THE RESULTS

The mean value of the exponent for the Caribou soils is 5.2; that for the Washburn soils, 5.93. Although all of the soils examined are acid, the Caribou soils are as a class, indeed almost without exception, more intensely acid than the Washburn soils. Of the general biological differences of the two types, the more interesting are those relating to the occurrence of potato scabs, since the growth of the causal organisms might be inhibited by the acidity of the Caribou soils but not by that of the Washburn soils. It has been shown for a number of microorganisms that growth is impossible beyond a certain intensity of acidity, as measured by the hydrogen-ion exponent. As compiled from various authorities by Clark and Lubs (4) the acid limit for six species studied lies above the acidity represented by the exponent 5.3. For a certain group of *Streptococci*, however, described by Ayers (1), the limiting exponents lie between 5.5 and 6.0, and without doubt other microorganisms will be found to fail to grow at such hydrogen-ion concentrations.

It is therefore not impossible that the relative freedom of soils of the Caribou type from disease may be due to the greater intensity of acidity.

SUMMARY

The Caribou loam and the Washburn loam from Aroostook County, Maine, possess before cultivation broadly different biological characteristics. Both types are largely devoted to potato culture, the Caribou loam being better adapted to this than the Washburn loam.

Cultivated soils of the Caribou loam type exhibit, when examined by the colorimetric method, considerably greater hydrogen-ion concentrations than do soils of the Washburn loam type. The average hydrogen-ion exponent for the Caribou loam was found to be 5.2; that of the Washburn loam 5.93.

The possibility is indicated that the relative freedom of the Caribou loam from potato scabs may be due to its greater hydrogen-ion concentration.

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FURTHER STUDIES OF THE NATURE OF AMMONIFICATION

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It has been proved by the author (1) that the process of ammonification and nitrification are autocatalytic chemical reactions and that the increase of ammonia and nitric acid in these processes are in accordance with the formula

$$\log \frac{x}{A-x} = K(t-t_1)$$

where x is the amount of nitrogen in the form of ammonia and of nitric acid which has been produced at the time t , A is the total amount of nitrogen in the form of ammonia and of nitric acid produced during the process, K is a constant, and t_1 is the time in which half of the total amount of nitrogen in the form of ammonia and of nitric acid is produced. This proof has been secured by the application of the autocatalytic chemical equation to the results of Lipman's, Lemmermann's and Warington's experiments. It will be both interesting and important, therefore, to test the proof of this subject further by personal experiments.

The present paper deals with the results of ammonification. The results of nitrification will be treated elsewhere later.

EXPERIMENTAL

Five different soils—Caribou silt loam, and Washburn silt loam from Maine, Superior clay from Wisconsin, Scottsburg silt loam from Indiana, and soil from the Arlington farm, Virginia, were selected and used for the experiment in an air-dried condition after having been passed through a 1-mm. sieve. Leucine and tyrosine were used as the chemicals to be ammonified. These were made in the laboratory by us from casein and from horn meal.

For the experiment 100 gm. of the soils and the chemicals equivalent to 100 mgm. of nitrogen were used. The amount of leucine and tyrosine used is, therefore, 0.9372 and 1.2943 gm., respectively. To the mixture 10 cc. of water was added and well mixed. The treated soils were kept in bottles. After incubation at a constant temperature (30°C.) for certain intervals of time as indicated in the following table, 10 gm. of the soils were taken and ammonia was determined by the usual distillation method with magnesia. In

¹ The investigations recorded in this paper were carried out in the laboratory of Soil Fertility Investigations, Bureau of Plant Industry, United States Department of Agriculture.

all cases the amount of ammonia obtained by distillation of the soil without addition of any compound, was subtracted from the quantity of ammonia found on distillation of the treated soils after they had undergone the process of ammonification. By weighing the bottles every morning we confirmed that the water content of the soils remained almost constant throughout the experiment.

The result obtained is as follows:

TABLE I

Milligrams of ammonia nitrogen actually produced from the compounds equivalent to 100 mgm. of nitrogen in 100 gm. of the soils and calculated by the equation given

1. Result with Arlington soil

a. Equation for leucine:

$$\log \frac{x}{78.46 - x} = 0.2032 (t - 6.85)$$

b. Equation for tyrosine:

$$\log \frac{x}{64.00 - x} = 0.1634 (t - 8.15)$$

DAYS	LEUCINE		TYROSINE	
	Calculated	Found	Calculated	Found
	mgm.	mgm.	mgm.	mgm.
2	7.35	7.77	5.76	3.12
5*	23.24	23.30	14.98	15.58
7	40.60	46.59	25.19	24.93
9*	57.43	62.12	37.07	37.39
12*	71.99	71.44	51.84	50.08
14	75.79	74.55	58.24	56.09
16*	77.38	77.66	61.17	61.21

2. Result with caribou soil

a. Equation for leucine:

$$\log \frac{x}{78.08 - x} = 0.1602 (t - 8.3)$$

b. Equation for tyrosine:

$$\log \frac{x}{71.62 - x} = 0.1816 (t - 8.3)$$

DAYS	LEUCINE		TYROSINE	
	Calculated	Found	Calculated	Found
	mgm.	mgm.	mgm.	mgm.
2	6.96	7.77	4.79	3.12
5*	17.83	17.20	14.39	12.80
7	29.85	29.95	26.31	26.71
9*	44.03	44.93	41.01	41.17
12*	62.19	59.02	59.04	57.87
14	69.55	67.12	65.58	65.11
16*	73.78	73.22	68.87	69.00

TABLE 1—Continued

3. Result with Indiana soil

a. Equation for leucine:

$$\log \frac{x}{84.53 - x} = 0.0992 (t - 8.55)$$

b. Equation for tyrosine:

$$\log \frac{x}{42.98 - x} = 0.1222 (t - 8.87)$$

DAYS	LEUCINE		TYROSINE	
	Calculated	Found	Calculated	Found
	mgm.	mgm.	mgm.	mgm.
2	15.47	8.32	5.43	3.34
5*	26.01	24.41	10.83	10.02
7	34.86	34.17	15.96	15.58
9*	44.44	44.37	21.88	21.81
12*	58.11	57.13	30.38	30.61
14	65.65	65.23	34.77	34.28
16*	71.49	71.44	37.89	38.40

4. Result with Superior soil

a. Equation for leucine:

$$\log \frac{x}{82.76 - x} = 0.1275 (t - 7.17)$$

b. Equation for tyrosine:

$$\log \frac{x}{58.36 - x} = 0.2045 (t - 5.35)$$

DAYS	LEUCINE		TYROSINE	
	Calculated	Found	Calculated	Found
	mgm.	mgm.	mgm.	mgm.
2	14.88	15.53	9.99	7.23
5*	28.63	24.85	27.87	26.15
7	40.34	40.38	39.77	40.51
9*	52.27	52.81	49.46	48.30
12*	66.62	65.23	55.92	54.53
14	72.93	74.55	57.39	56.20
16*	77.02	77.66	58.14	57.65

5. Result with Washburn soil

a. Equation for leucine:

$$\log \frac{x}{82.74 - x} = 0.201 (t - 4.3)$$

b. Equation for tyrosine:

$$\log \frac{x}{79.56 - x} = 0.0916 (t - 5.75)$$

DAYS	LEUCINE		TYROSINE	
	Calculated	Found	Calculated	Found
	mgm.	mgm.	mgm.	mgm.
2	21.14	10.87	24.82	12.46
5*	48.03	48.15	36.64	35.84
7	64.31	69.89	44.96	45.19
9*	74.31	76.20	52.90	51.42
12*	80.44	79.20	62.76	61.21
14	81.82	80.76	67.68	66.22
16*	82.37	82.32	71.34	70.12

* These points were used for the calculation of value A.

It will be seen from the table that the observed and calculated values of x are in good agreement. There is, therefore, no doubt that the process of ammonification is an autocatalytic chemical reaction and that the increase of ammonia in this process is in accordance with the formula $\log \frac{x}{A-x} = K(t - t_0)$, as stated in the previous paper.

From the practical standpoint, it might be considered that while the ammonification of a compound is still going on in the soil, a part of the ammonia produced is oxidized to nitrites and nitrates. The loss of nitrogen in the form of ammonia is also caused through denitrification. For such reasons, the ratio between the nitrogen in the compound added and the nitrogen in the ammonia produced never attains its final value. The total amount of nitrogen in the compound added, consequently, might be gradually transformed into nitrogen of ammonia form. But when we consider ammonification itself from the scientific standpoint, it is very interesting and important to know whether the total amount of nitrogen added will be ammonified. The rate of transformation of nitrogen in the process of ammonification will be clearly shown by the equation, because A in the equation denotes the maximum amount of nitrogen produced in the form of ammonia. The actual value of A is, however, always less than the amount of nitrogen added. This means that the nitrogen added is not totally transformed into the nitrogen in the form of ammonia in the process of ammonification. For the sake of convenience we have brought together our results in table 2.

TABLE 2
Rate of transformation of nitrogen in the process of ammonification

SOILS	LEUCINE		TYROSINE	
	Nitrogen added mgm.	Value of A mgm.	Nitrogen added mgm.	Value of A mgm.
Arlington.....	100.00	78.46	100.00	64.00
Caribou.....	100.00	78.08	100.00	71.62
Indiana.....	100.00	84.53	100.00	42.98
Superior.....	100.00	82.76	100.00	58.36
Washburn.....	100.00	82.74	100.00	79.56

The rate of transformation seems to be dependent upon the conditions of the experiment as to soils, chemicals, moisture, temperature, etc. In our experiment, the value A for leucine was always greater than that for tyrosine, that is, leucine ammonified to a greater extent than did tyrosine. The value A of each compound was also greatly influenced by the soils.

SUMMARY

1. We have confirmed again that the process of ammonification is an auto-catalytic chemical reaction and that the increase of ammonia in the process is in accordance with the formula:

$$\log \frac{x}{A-x} = K(t - t_1).$$

2. The total amount of nitrogen added to be ammonified does not transform into the nitrogen in the form of ammonia in the process of ammonification.

3. The amount of nitrogen transformed into ammonia nitrogen is greatly influenced by both the soils and chemical compounds used.

In conclusion, it is a pleasure for the author to acknowledge his indebtedness to Dr. Oswald Schreiner and Dr. Elbert C. Lathrop, of the Bureau of Plant Industry, United States Department of Agriculture.

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NITRIFICATION AS A MEASURE OF THE AVAILABILITY OF DIFFERENT FORMS OF CALCIUM CARBONATE WHEN EMPLOYED AS CORRECTORS OF SOIL ACIDITY

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Last July a sample of finely-ground, soft coral limestone was submitted to this laboratory for analysis by one of the local lime companies. A request for our opinion of the material as a corrector of soil acidity accompanied the sample. The analysis of this limestone appears in table 1.

TABLE 1
Analysis of soft coral limestone

	<i>per cent</i>
Moisture.....	1.2
Volatile matter (chiefly CO ₂).....	41.2
SiO ₂ , Al ₂ O ₃ , Fe ₂ O ₃	2.4
Lime (CaO).....	52.6
Magnesia (MgO).....	1.3
Phosphorus pentoxide (P ₂ O ₅).....	0.1
Sulfur trioxide (SO ₃).....	0.6
Lime calculated as carbonate.....	93.6

After receiving the above report the company in question desired us to make a few tests, if possible to ascertain the solubility or availability of this finely-ground product as compared with coral sand which at present is being quite generally applied to soils in the acid districts. Thinking this to be a subject of some importance to the plantations located on the acid soils of Hawaii, as well as a subject of some scientific interest, a series of experiments were planned to test out the relative rates of availability of the two forms of lime carbonate in question. The results of these experiments form an excuse for the publication of this article.

There are two points to be recognized and remembered in reading this report. First, coral sand and finely ground coral limestone are identical chemically—their only difference lies in mechanical condition, i.e., degree of fineness and hardness of grain. Second, in applying lime to sugar cane lands for the correction of soil acidity we do it, not so much to benefit the cane plant directly (for we know that, if properly fertilized, cane can be grown on very acid soils), as to enhance beneficial bacterial activity. If this is accomplished, available plant-food is at all times being prepared, resulting in a more normal feeding of the cane, and eventually in increased yields.

At first this problem of lime availability appeared somewhat puzzling, for time did not permit of extended vegetative tests with growing plants. Furthermore, it is obviously impossible to add the materials to exact neutrality in an acid soil and determine the rates of availability by the ordinary lime requirement methods, for, if correct amounts of the materials were added upon such chemical analysis a neutral soil would at all times result, irrespective of the size of the calcium carbonate particles. In fact, the Veitch method was tried which showed the futility of such a method of attack. It then occurred to the writer that, while nitrification is low in an acid soil, it picks up with remarkable celerity upon the addition of lime carbonate *in readily available forms*. On the other hand, if lime carbonate is added in chunks or in unavailable forms, as in hard shells or coarse marble fragments, nitrification would be enhanced but slightly.

The plan of the investigation follows.

About 100 pounds of a very acid surface soil was secured, through the courtesy of Mr. W. P. Naquin, from one of the upper fields of the Pacific Sugar Mill at Kukuihaele, Hawaii. It is a reddish brown clay loam of considerable potential fertility and previous experiments had shown it to contain a nitrifying flora which awaited only a proper lime application to render it vigorously active. An analysis of this soil is given in table 2.

TABLE 2
Analysis of Kukuihaele Mauka surface soil

Agricultural method of analysis (1)

	per cent
Phosphoric acid (P_2O_5).....	0.88
Potash (K_2O).....	0.33
Lime (CaO).....	0.31
Total nitrogen.....	0.44

Citrate-soluble analysis [Dyer's modified method] (1)

Phosphoric acid (P_2O_5).....	0.0018
Potash (K_2O).....	0.0283
Lime (CaO).....	0.0560

Lime-requirement (Veitch method) 1.068 per cent lime carbonate (16 tons per acre-foot).

This soil, as it exists in the field, is of good physical condition but underlain at a depth of about 6 inches by a heavy, hard subsoil of somewhat lighter color.

The bag of soil, upon its arrival at the laboratory, was air-dried in the shade, sifted through a 3-mm. sieve and thoroughly mixed. Two hundred and sixteen 100-gm. portions were now weighed out into clean, sterile glass tumblers and covered with glass Petri dishes. These were arranged in 9 series of 12 duplicate soil cultures each. The treatment of each series was identical, being as follows:

NO.	TREATMENT
1- 1A	Blank on soil, exactly neutralized with coral sand
2- 2A	Blank on soil, exactly neutralized with ground limestone
3- 3A	0.3 gm. dried blood added (40.5 mgm. N)
4- 4A	0.3 gm. dried blood neutralized with coral sand
5- 5A	0.3 gm. dried blood twice neutralized with coral sand
6- 6A	0.2 gm. ammonium sulfate added (42.4 mgm. N)
7- 7A	0.2 gm. ammonium neutralized with coral sand
8- 8A	0.2 gm. ammonium twice neutralized with coral sand
9- 9A	0.3 gm. dried blood added, neutralized with ground limestone
10-10A	0.3 gm. dried blood added, twice neutralized with ground limestone
11-11A	0.2 gm. ammonium sulfate added, neutralized with ground limestone
12-12A	0.2 gm. ammonium sulfate added, twice neutralized with ground limestone

A brief explanation of these treatments may not be superfluous. The coral sand employed was of very good quality. Only that portion between 0.5 mm. and 1mm. in size was used. This treatment eliminated all of the larger grains of sand, shells, sticks, pieces of seaweed, etc. Upon analysis it was found to contain 89 per cent of lime carbonate. An analysis of the finely-ground coral limestone appears in table 1. In supplying both of these materials to the cultures, allowances were made for contained impurities so that equal and correct percentages of *lime carbonate* (CaCO_3) were added in all cases to neutralize the soil acidity exactly, or double this amount, as the case might be.

In soil cultures 1 and 1A no nitrogen was added. Enough coral sand was supplied to neutralize the soil acidity exactly.

Soil cultures 2 and 2A were treated in exactly the same way as no. 1 and 1A, except that ground limestone was here substituted for the coral sand.

Soil cultures 3 and 3A received 0.3 gm., of dried blood only (40.5 mgm. organic nitrogen).

Soil cultures 4 and 4A were exactly the same as no. 3 and 3A, except that the soil acidity was here exactly neutralized with coral sand.

Soil cultures 5 and 5A were the same as no. 3 and 3A, with the exception that here twice as much coral sand as required was added.

Soil cultures 6 and 6A received 0.2 gm. of ammonium sulfate only (42.4 mgm. ammonia nitrogen).

Soil cultures 7 and 7A were the same as no. 6 and 6A, except that here enough coral sand to neutralize soil acidity exactly was added.

Soil cultures 8 and 8A were identical with no. 6 and 6A, except that twice as much coral sand was added as was actually needed.

Soil cultures 9 and 9A received 0.3 gm. of dried blood and exactly enough ground coral limestone to neutralize the soil acidity.

Soil cultures 10 and 10A were similar to no. 9 and 9A, except that here twice as much ground limestone was added.

Soil cultures 11 and 11A received 0.2 gm. of ammonium sulfate each after being exactly neutralized with ground limestone.

Soil cultures 12 and 12A were the same as no. 11 and 11A except that twice as much ground limestone was added.

To all of the soil cultures optimum amounts of sterile distilled water were added. The incubation temperature was 28°C. Two cubic centimeters of water were added each week to each culture to make up for loss from evapo-

TABLE
Availability of coral sand and ground limestone

SOIL CULTURE NO.	TREATMENTS	PERIOD OF INCUBATION							
		2 weeks Series I		4 weeks Series II		6 weeks Series III		8 weeks Series IV	
		N nitrified per culture	Nitrogen nitrified						
1	Blank + sand.	4.00	0.91	4.00	0.91	4.50	1.02	9.00	2.04
2	Blank + ground limestone.	4.80	1.09	4.50	1.02	6.00	1.36	9.00	2.0
3	Dried blood only.	4.80	11.8	4.20	10.4	7.50	18.5	12.00	29.6
4	Dried blood + sand.	9.00	22.2	10.00	24.7	12.00	29.6	18.00	44.4
5	Dried blood + 2 sand.	10.40	25.7	11.00	27.2	16.00	39.5	21.00	51.9
6	Ammonium sulfate only.	3.40	8.0	4.80	11.3	8.00	18.9	7.50	17.7
7	Ammonium sulfate + sand.	10.00	23.6	28.00	66.0	27.00	63.7	29.50	69.6
8	Ammonium sulfate + 2 sand.	15.00	35.4	30.00	70.7	28.00	66.0	33.00	77.5
9	Dried blood + ground limestone.	12.00	29.6	25.00	61.7	25.00	61.7	25.00	61.7
10	Dried blood + 2 ground limestone.	13.00	32.1	25.00	61.7	26.00	64.2	25.00	61.7
11	Ammonium sulfate + ground limestone	13.00	30.7	44.50	105.0	45.00	106.1	43.00	101.4
12	Ammonium sulfate + 2 ground limestone	32.00	75.5	45.00	106.1	45.00	106.1	44.00	103.5
Blank	Blank on soil, no additions. Incubated 20 weeks								

¹ These are averages of closely agreeing duplicate determinations.

tion. All nine series (216 cultures) were started together on August 7, 1916.

As the chief object of the experiment was to note the progressive effect of the different forms of lime carbonate on the nitrification of organic nitrogen and sulfate nitrogen, one series was dried out and analyzed for nitrates at the end of each period of 2 weeks during the first 16 weeks. The last, series IX, was allowed to incubate 5 months (20 weeks).

Table 3 imparts all of the data obtained, while figure 1 graphically presents these same results. It should be stated that nitrates were determined in these cultures by the modified phenoldisulphonic acid method of Gill (2), calcium carbonate (3) being used as the flocculating agent.

Before discussing the data here presented it should be stated that, under mainland conditions according to data previously secured by the writer and by others, the amounts of nitrate nitrogen biologically formed during one

3

coral limestone as measured by nitrification¹

BATION OF CULTURES										GENERAL AVERAGES OF ALL SERIES	
10 weeks Series V		12 weeks Series VI		14 weeks Series VII		16 weeks Series VIII		20 weeks Series IX		Nitrified per culture	Nitrogen nitrified
N nitrified per culture	Nitrogen nitrified	mgm.	per cent								
12.00	2.73	9.00	2.04	10.00	2.27	8.00	1.82	9.00	2.04	7.72	1.75
12.00	2.73	10.00	2.27	11.00	2.50	7.50	1.70	9.00	2.04	8.20	1.86
18.00	44.4	16.00	39.5	13.00	32.1	9.00	22.2	11.00	27.2	10.50	25.9
20.00	49.4	22.00	54.3	25.00	61.7	20.00	49.4	20.00	49.4	17.33	42.8
24.00	59.3	27.00	66.7	25.00	61.7	22.00	54.3	28.00	69.1	20.50	50.6
8.00	18.9	7.00	16.5	6.00	14.1	6.00	14.1	7.50	17.7	6.46	15.2
30.00	70.7	30.00	70.7	31.00	73.1	24.00	56.6	22.00	51.9	25.65	60.5
32.00	75.5	32.00	75.5	32.00	75.5	25.00	59.0	30.00	70.7	28.55	67.3
26.00	64.2	26.00	64.2	26.00	64.2	25.00	61.7	28.00	69.1	24.62	60.8
30.00	74.1	29.00	71.6	28.00	69.1	lost		29.00	71.6	25.62	63.3
44.00	103.8	44.00	103.8	40.00	94.3	40.00	94.3	39.00	92.0	39.16	92.4
45.00	106.1	44.00	103.8	43.00	101.4	41.00	96.7	40.00	94.3	42.11	99.3
								1.60	0.36	1.60	0.36

month's incubation period in soil cultures under optimum conditions of moisture and temperature (in an incubator) are very nearly equivalent to the amounts formed in soils in the field during a period of from 2 to 3 months.¹ Therefore, the "Incubation Period in Weeks" as shown in figure 1 should in

¹ Experiments testing out this point for Hawaiian soils under Hawaiian climatical conditions are now being instigated at this experiment station.

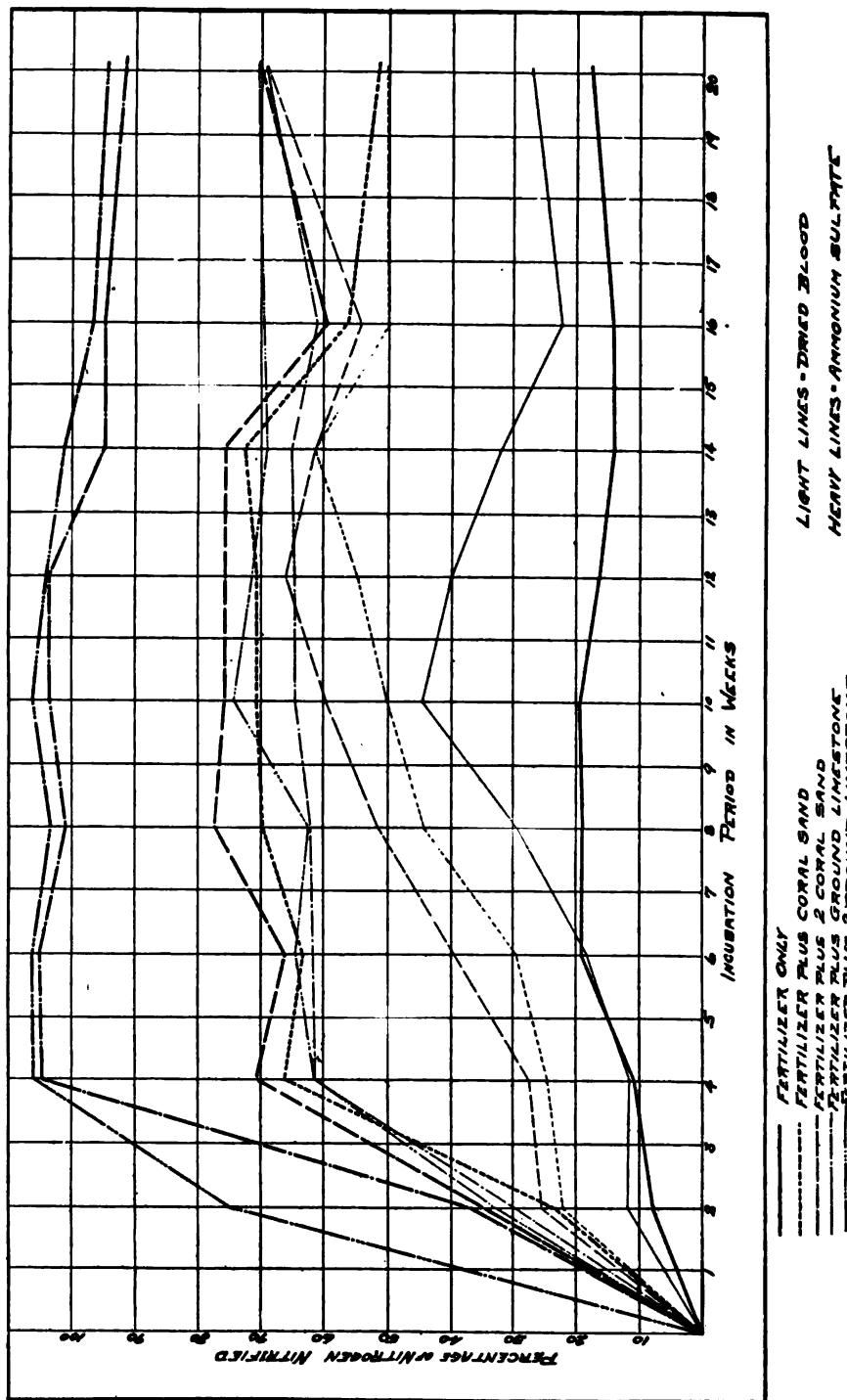


FIG. 1. CURVES SHOWING EFFECT OF CORAL SAND AND FINELY-GROUND CORAL LIMESTONE ON NITRIFICATION

all probability be multiplied by about 2 or 3 in each case if approximate field conditions are desired.

A number of very interesting facts are shown by a study of the data and graphs presented. Probably the most striking is the effect of the finely-ground coral limestone on the *rate* of nitrification. This is especially true where ammonium sulfate was used as the source of nitrogen (see heavy lines in figure 1). At the end of 2 weeks, only 8 per cent of the sulfate nitrogen was transformed where no lime was applied. Where exactly enough ground limestone to neutralize soil acidity was used, nearly 31 per cent was nitrified, and where twice this amount was added, 75 per cent of the sulfate nitrogen was converted into nitrate in this *previously acid* soil. The same general trend is noted where coral sand instead of ground limestone was used, but the increments of gain over the cultures without lime are here a little less than half the above.

At the end of one month, where ground limestone was used, all of the added sulfate nitrogen was nitrified, as well as a small amount of the soil's own organic nitrogen, while where coral sand was added only about 70 per cent of the added nitrogen was transformed. At no period of time after this was coral sand, either to exact neutrality or in twice this amount, able to effect a nitrification of much over 75 per cent of the sulfate nitrogen originally added. On the other hand, ground limestone, from the fourth to the fifteenth week, in all cases affected a complete nitrification of all added sulfate nitrogen. It will be noted that after about the fifteenth week there was a gradual dropping off of nitrate in all of the cultures. This is doubtless due in part to denitrification, in part to the complete use of all bases present by the nitrous and nitric acids formed by the nitrifying bacteria, and in part to the formation of other bacterial products which were inimical to continued active bacterial growth. As will also be seen, the additions of twice the required amounts of lime (either as ground limestone or as coral sand), had but slight advantages over those quantities needed to neutralize soil acidity exactly. The only pronounced exception is with the ground limestone at the start of the incubation period. Here an excess of lime appears to have accelerated considerably the initial bacterial action.

Where dried blood was substituted for ammonium sulfate as the carrier of nitrogen, the same general tendencies as above noted occurred (see light lines in figure 1), although the absolute percentages of added nitrogen nitrified are less. At the end of 4 weeks ground limestone effected over three times as much nitrification (over the no lime cultures) as did coral sand, but this ratio gradually narrowed with prolonged incubation until, at the end of 14 weeks, the coral sand was to the ground limestone as 8 is to 9. Continued incubation from this point had little effect on nitrate formation where sand was added, but there continued to be a slight increase with the finely-ground limestone. Double the required amounts of coral sand (where dried-blood nitrogen was used) had a far greater enhancing effect on nitrification than similar double

portions of the ground limestone, for at the end of 12 weeks twice the former gave approximately the same amount of nitrate formed as once the latter. The reason for this is probably to be found in the larger amount of *fine* material added in a double application of the coral sand, for, as this work clearly shows, the finer the material the more readily it corrects acidity.

The above discussion has been based on the *percentage* of added nitrogen transformed. Let us here briefly consider the *absolute amounts* of nitrate produced. It will be remembered that the same soil was used throughout, also that approximately equal amounts of nitrogen were in all cases added (40.5 mgm. as blood nitrogen and 42.4 mgm. as ammonium sulfate nitrogen). Table 3, shows definitely that, where no lime in any form is added, larger quantities of nitrogen are converted into nitrate from blood nitrogen than from sulfate nitrogen, but where *lime* of any description is added the *absolute reverse* is true. This is readily explained. Where ammonium sulfate only is applied, the basic ammonium radicle (NH_4) is in part converted into nitrate, leaving the acid sulfate radicle (SO_4) free. The latter, combining with water (H_2O), immediately forms sulfuric acid (H_2SO_4). Thus, as slow nitrification progresses the soil becomes more and more acid until, as shown in figure 1, at the end of 6 weeks such an excess of acid has accumulated that from then on nitrification gradually falls off to the end of the experiment. On the other hand, when blood alone is applied, vigorous ammonification at once sets in with an evolution of far more basic ammonium carbonate than the sluggish nitrifying bacteria can oxidize. The result is that much of the soil acidity is at once neutralized, which in turn renders conditions more favorable to active nitrification. Thus *at first* the nitrification of blood is fairly rapid (fig. 1), but a time eventually comes when all of this basic ammonium carbonate is either nitrified or neutralized (at the end of 10 weeks' incubation period). From here on will be noticed a very rapid falling off in the amounts of nitrate formed, which continues to the end of the experiment. Stated concisely, organic forms of nitrogen are always more quickly and readily nitrified in acid soils than are mineral ammonium salts.

A few valuable facts are gleaned from a study of the figures presented in table 3 which are impossible of graphic representation in figure 1.

When we consider the soil's own organic nitrogen (no additions of either dried blood or ammonium sulfate) we find that it makes practically no difference as to absolute amounts of nitrogen nitrified whether coral sand or finely ground limestone is used, although slightly quicker action is secured from applications of the latter. Averaging all of the figures throughout the 20 weeks' test we have, where sand was applied, 1.75 per cent of the soil's nitrogen nitrified; where ground limestone was used, 1.86 per cent, and where no lime was employed 0.36 per cent. The reason for the close approach of sand to ground limestone where the soil's own organic nitrogen was concerned is probably to be found in the fact that here such *small* amounts of nitric acid are produced that enough very fine material is present in the sand added to neu-

tralize it, as well as the more active soil acidity. Experiments pertaining to the effect of *partially* neutralizing soil acidity on the nitrification of the soil's own organic nitrogen, sulfate nitrogen and blood nitrogen, are now in progress at this station.

Considering absolute amounts of nitrogen made available (transformed to nitrate), it is noteworthy that an application of the required amount of lime to this soil *without added nitrogen* supplies *more nitrate* to the growing cane through the nitrification of the soil's own organic nitrogen than an application of $1\frac{1}{2}$ tons per acre of ammonium sulfate *without lime*, and four-fifths as much as an application of 1 ton of dried blood *without lime*, provided moisture and temperature conditions are suitable. In other words, lime and no fertilizer (except nitrates) could here pay better than fertilizer and no lime.

CONCLUSIONS

Taking the average figures for each 2-week incubation period throughout the 20 weeks (see last two columns of figures in table 3) the following conclusions may be drawn:

1. Where no additions of nitrogen are made, coral sand and ground coral limestone are about equally effective in enhancing the nitrification of an acid soil's own organic nitrogen. After neutralizing Hawaii acid soils, the average amounts of nitrate formed over a period of 5 months under optimum conditions are comparatively small.

2. The increment of gain in nitrate formed over the soil exactly neutralized, due to adding twice the amounts of lime required (either as coral sand or as ground limestone), is too slight to warrant double applications. Twice the required amounts of coral sand effect a greater increase in nitrate produced over the soils exactly neutralized than do twice the amounts of finely-ground coral limestone.

3. Where *coral sand* was used in sufficient amounts to bring the soils to exact neutrality, the following percentages of gain over the soils to which no lime in any form was added, are indicated:

	<i>per cent</i>
No nitrogen added (soil's own nitrogen).....	486
Dried blood nitrogen added.....	165
Ammonium sulfate nitrogen added.....	398

4. Where *finely-ground coral limestone* was used to neutrality, the following percentages of gain over the "no lime" cultures are indicated:

	<i>per cent</i>
No nitrogen added (soil's own nitrogen).....	518
Dried blood nitrogen added.....	235
Ammonium sulfate nitrogen added.....	608

5. Comparing the average percentage of increased nitrate production, due to neutralizing exactly the soil with coral sand, with that brought about by

the addition of finely-ground coral limestone, where both ammonium sulfate nitrogen and dried blood nitrogen were supplied, we have, as a general average over the entire incubation period of 5 months, an increase of 281 per cent due to sand applications and an increase of 421 per cent due to ground limestone applications. From these figures a simple ratio shows that, when lime availability is measured in the soil by enhanced nitrification, 1 ton of the finely-ground limestone is practically equivalent, in neutralizing soil acidity, to 1.5 tons of the best grade of coral sand.

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VEGETATION EXPERIMENTS ON THE AVAILABILITY OF TREATED PHOSPHATES

JACOB G. LIPMAN AND HARRY C. McLEAN

New Jersey Agricultural Experiment Stations

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The authors of this paper suggested in an earlier publication¹ the use of sulfur as a means for making available the phosphorus in ground phosphate rock. They showed that ground sulfur may be used successfully for this purpose. Experiments conducted since then have confirmed the earlier results and will be reported at a later date. It is the purpose of the authors to record in this paper the results secured in vegetation experiments from ground phosphate rock otherwise untreated, from ground phosphate rock previously composted with sulfur, and from acid phosphate. In arranging for these vegetation experiments, it was felt that the availability of the phosphate rock previously composted with sulfur should be demonstrated not merely by chemical tests, but also by actual tests with plants. Accordingly, quantities of ground phosphate rock (floats), ground phosphate rock previously composted with sulfur, and acid phosphate, containing equivalent amounts of phosphorus, were used in the pot experiments described in the following pages. The experiments were carried out by means of glazed earthenware pots of 1 gallon capacity. In the series in which cabbage was grown, however, 2-gallon pots were used. In each pot there was placed 10 pounds of white quartz sand and, in addition, 5 gm. of calcium carbonate, 1 gm. of potassium sulfate, 0.25 gm. of magnesium sulfate, 0.12 gm. of ferric sulfate and 1.5 gm. of sodium nitrate. In order to eliminate sulfur as a limiting factor, pots 1, 2, 3 and 4 in each series received additions of calcium sulfate equivalent in amount to that contained in the sulfur-floats mixture. The additions described above provided, therefore, all of the essential plant-food constituents except phosphorus. The special treatment consisted of applications of no phosphorus in pots 1 and 2 of each series; of ground phosphate rock in pots 3 and 4 of each series; of ground phosphate rock and sulfur previously composted in pots 5 and 6 of each series, and of acid phosphate in pots 7 and 8 of each series. The amount of phosphorus actually furnished was equivalent in each case to that contained in 1 gm. of floats. The fertilizer materials were added to the 10 pounds of sand used in each pot and thoroughly mixed with the sand in a flat pan before they were transferred to the pots. However, in the series in which cabbage was grown,

¹ Lipman, J. G., McLean, H. C., and Lint, H. C., 1916. Sulfur oxidation in soils and its effect on the availability of mineral phosphates. *In* Soil Sci., vol. 2, no. 6, p. 499-538.

20 pounds of sand and double quantities of the different fertilizer materials were employed. There were, in all, 11 series of 8 pots each. The crops grown in the tests included cabbage, rape, buckwheat, soybeans, corn, sorghum, barley, millet, turnips, wax beans and alfalfa. The plants grown in each series were thinned out to leave the same number of plants per pot. The number of these varied with the crop grown. The moisture content was maintained during the experiment at about 10 per cent.

It was observed soon after germination that the plants in the pots which received no additions of phosphorus, and in those which received additions of floats, made very slight growth. On the other hand, the plants in the pots which had received additions of floats previously composted with sulfur, and in those treated with acid phosphate, made very good growth. After several weeks the crops were photographed and harvested. After thorough drying, records were made of the dry weights of the plants harvested from each pot. These weights are recorded in table 1. After the first crop was harvested, the sand in each pot was thoroughly stirred with a trowel and a second application of 1 gm. of sodium nitrate was made in order to preclude the possibility of nitrogen becoming the limiting factor in production. Without further treatment a residual crop was grown in all but three of the series. The residual crops were later harvested, dried and their weights recorded. A record also of these weights appears in table 1. Photographs showing the appearance of the plants in three of the series are given in plate 1.

An examination of the table will show that the different crops showed marked variations in their ability to utilize the phosphorus in the floats. It is particularly interesting to note that buckwheat was able to make very good growth in pots 3 and 4 in which floats were the only source of phosphorus. Also, cabbage and rape were able to utilize the phosphorus of floats to a fairly marked extent. On the other hand, turnips, barley, millet, soybeans, wax beans, alfalfa, corn and sorghum were unable to utilize the phosphorus in floats to an appreciable extent. These results indicate a practical application in that floats, now unquestionably the cheapest source of phosphorus, might be used for the growing of buckwheat, the latter to be turned under as a green manure. In this manner, buckwheat might be used as a means for supplying phosphorus at a low cost to small grains, corn and some of the legumes.

It might also be noted that the floats previously composted with sulfur became a very satisfactory source of phosphorus. This was, of course, expected, since analyses of the composted mixtures of floats and sulfur showed that in a period of 30 weeks most of the phosphorus in the floats had become available, according to the method of the Association of Official Agricultural Chemists. It will be observed that, in the case of the rape, turnips, buckwheat, barley, millet, soybeans, wax beans and sorghum, the floats-sulfur compost was as effective as the acid phosphate. Where these crops were followed by residual crops, viz., millet, corn and sorghum, the floats-sulfur compost also showed up to good advantage. On the other hand, in the case

TABLE I
The influence of treated phosphates on the yields of dry matter

POT NUMBER	TREATMENT	FIRST CROP			RESIDUAL CROP			TOTAL INCREASE OVER CHECK —BOTH CROPS
		Weight of crop	Average	Increase over check	Weight of crop	Average	Increase over check	
Cabbage								
1	Check (no phosphorus).....	1.50			3.40			
2	Check (no phosphorus).....	1.10	1.30		4.00	3.70		
3	Raw rock phosphate.....	4.25			4.20			
4	Raw rock phosphate.....	3.40	3.83	2.53	3.75	3.98	0.28	2.81
5	Compost.....	9.35			17.00			
6	Compost.....	6.30	7.83	6.53	19.40	18.20	14.50	21.03
7	Commercial acid phosphate..	11.55			39.25			
8	Commercial acid phosphate..	13.03	12.29	10.99	40.75	40.05	36.35	47.34
Rape								
1	Check (no phosphorus).....	1.20			0.20			
2	Check (no phosphorus).....	4.90	3.05		0.85	0.53		
3	Raw rock phosphate.....	3.85			0.35			
4	Raw rock phosphate.....	6.70	5.28	2.23	1.25	0.80	0.27	2.50
5	Compost.....	8.50			9.20			
6	Compost.....	8.90	8.70	5.65	8.15	8.68	8.15	13.80
7	Commercial acid phosphate..	8.46			9.85			
8	Commercial acid phosphate..	8.35	8.41	5.36	10.00	9.93	9.40	14.76
Turnips								
1	Check (no phosphorus).....	0.25			0.40			
2	Check (no phosphorus).....	0.25	0.25		0.40	0.40		
3	Raw rock phosphate.....	0.70			0.60			
4	Raw rock phosphate.....	0.50	0.60	0.35	0.60	0.60	0.20	0.55
5	Compost	9.75			7.50			
6	Compost	10.40	10.08	9.83	7.25	7.38	6.98	16.81
7	Commercial acid phosphate..	10.80			9.00			
8	Commercial acid phosphate..	9.25	10.03	9.78	9.00	9.00	8.60	18.38
Buckwheat								
1	Check (no phosphorus).....	5.20			0.30			
2	Check (no phosphorus).....	5.40	5.30		0.25	0.28		
3	Raw rock phosphate.....	12.20			2.85			
4	Raw rock phosphate.....	12.90	12.55	7.25	1.60	2.23	1.95	9.20
5	Compost.....	13.40			8.55			
6	Compost.....	13.80	13.60	8.30	10.25	9.40	9.12	17.42
7	Commercial acid phosphate..	14.00			11.50			
8	Commercial acid phosphate..	14.80	14.40	9.10	10.75	11.13	10.85	19.95

TABLE 1—(Continued)

POT NUMBER	TREATMENT	FIRST CROP			RESIDUAL CROP			TOTAL INCREASE OVER CHECK —BOTH CROPS
		Weight of crop	Average	Increase over check	Weight of crop	Average	Increase over check	
		grams	grams	grams	grams	grams	grams	
Barley								
1	Check (no phosphorus).....	1.05			3.50			
2	Check (no phosphorus).....	1.10	1.08		2.75	3.13		
3	Raw rock phosphate.....	1.55			3.20			
4	Raw rock phosphate.....	1.25	1.40	0.32	3.15	3.18	0.05	0.37
5	Compost.....	7.60			6.25			
6	Compost.....	8.05	7.83	6.75	4.25	5.25	2.12	8.87
7	Commercial acid phosphate..	8.00			6.80			
8	Commercial acid phosphate..	7.80	7.90	6.82	5.70	6.25	3.12	9.94
Millet								
Sorghum								
1	Check (no phosphorus).....	0.35			0.65			
2	Check (no phosphorus).....	0.45	0.40		0.75	0.70		
3	Raw rock phosphate	0.55			0.90			
4	Raw rock phosphate	0.60	0.58	0.18	0.90	0.90	0.20	0.38
5	Compost.....	16.00			2.75			
6	Compost.....	14.55	15.28	14.88	1.50	2.13	1.43	16.31
7	Commercial acid phosphate..	15.20			2.10			
8	Commercial acid phosphate..	15.30	15.25	14.85	0.75	1.43	0.73	15.58
Soy Beans								
Corn								
1	Check (no phosphorus).....	9.80			3.08			
2	Check (no phosphorus).....	10.15	10.48		4.60	3.84		
3	Raw rock phosphate	8.90			4.35			
4	Raw rock phosphate	10.95	9.93	(-0.55)	3.95	4.15	0.31	(-0.24)
5	Compost.....	27.70			10.35			
6	Compost.....	25.50	26.60	16.12	12.15	11.25	7.41	23.53
7	Commercial acid phosphate..	24.10			11.20			
8	Commercial acid phosphate..	24.50	24.30	13.82	12.30	11.75	7.91	21.73
Wax Beans								
Millet								
1	Check (no phosphorus).....	7.70			0.35			
2	Check (no phosphorus).....	7.50	7.60		0.35	0.35		
3	Raw rock phosphate	6.70			0.85			
4	Raw rock phosphate	9.10	7.90	0.30	0.65	0.75	0.40	0.70
5	Compost.....	12.20			7.00			
6	Compost.....	12.60	12.40	4.80	4.00	5.50	5.15	9.95
7	Commercial acid phosphate..	13.10			6.00			
8	Commercial acid phosphate..	12.30	12.70	5.10	6.25	6.13	5.78	10.88

TABLE 1—(Concluded)

POT NUMBER	TREATMENT	FIRST CROP			RESIDUAL CROP			TOTAL INCREASE OVER CHECK —BOTH CROPS
		Weight of crop	Average	Increase over check	Weight of crop	Average	Increase over check	
Alfalfa								
1	Check (no phosphorus).....	0.20						
2	Check (no phosphorus).....	0.15	0.18					
3	Raw rock phosphate.....	0.18						
4	Raw rock phosphate.....	0.15	0.17	(-0.01)				
5	Compost.....	1.80						
6	Compost.....	1.75	1.78	1.60				
7	Commercial acid phosphate.....	4.30						
8	Commercial acid phosphate.....	4.30	4.30	3.12				
Corn								
1	Check (no phosphorus).....	3.00						
2	Check (no phosphorus).....	2.40	2.70					
3	Raw rock phosphate.....	3.35						
4	Raw rock phosphate.....	2.80	3.08	0.38				
5	Compost.....	10.70						
6	Compost.....	11.50	11.10	8.40				
7	Commercial acid phosphate.....	14.80						
8	Commercial acid phosphate.....	13.60	14.20	11.50				
Sorghum								
1	Check (no phosphorus).....	0.70						
2	Check (no phosphorus).....	0.40	0.55					
3	Raw rock phosphate.....	1.20						
4	Raw rock phosphate.....	0.90	1.05	0.50				
5	Compost.....	9.90						
6	Compost.....	12.25	11.08	10.53				
7	Commercial acid phosphate.....	12.20						
8	Commercial acid phosphate.....	10.90	11.55	11.00				

of cabbage followed by corn, the acid phosphate was distinctly superior to the floats-sulfur compost. The discrepancy in this particular series is difficult to account for and will be made the subject of further study. Meanwhile, it is sufficient to state in this preliminary paper that floats properly composted with ground sulfur may become a source of available phosphorus, as shown both by chemical analyses and by vegetation tests.

SUMMARY

1. It has been shown in this paper that some crops can utilize the phosphorus in floats to good advantage. This is particularly true of buckwheat.
2. The ability of buckwheat to use effectively the phosphorus of ground, but otherwise untreated, phosphate rock suggests that this crop may be made a

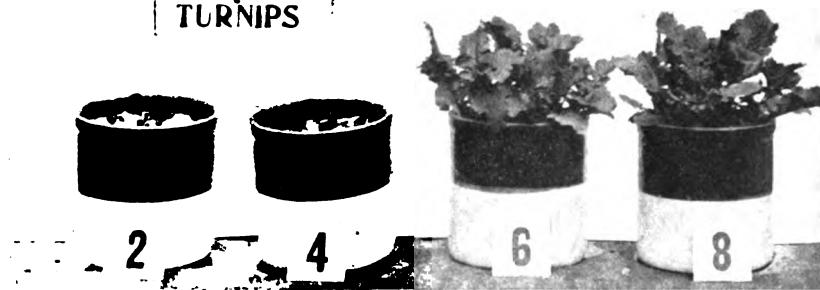
valuable green manure and employed to increase the content in the soil of organic matter and of available phosphorus.

3. Ground phosphate rock properly composted with ground sulfur, becomes a source of available phosphorus and may be employed to advantage as a substitute for acid phosphate.

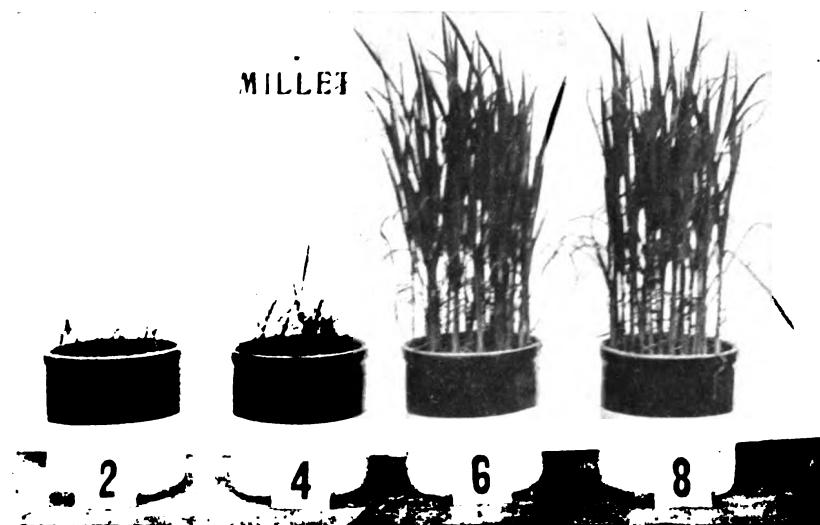
PLATE 1

Pot 2, no phosphorus; pot 4, floats; pot 6, compost; pot 8, commercial acid phosphate.

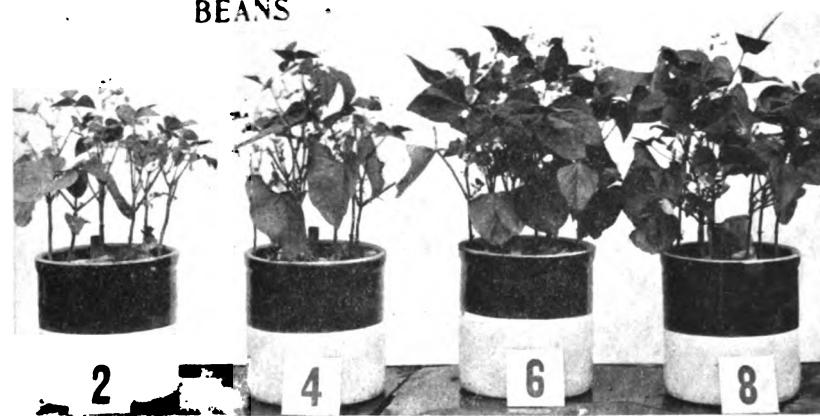
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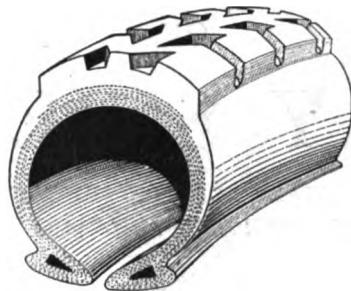
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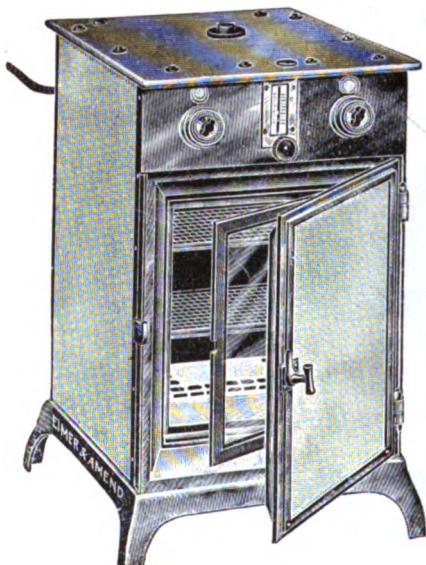
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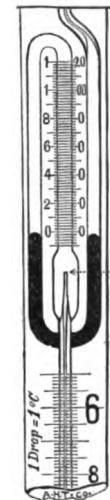
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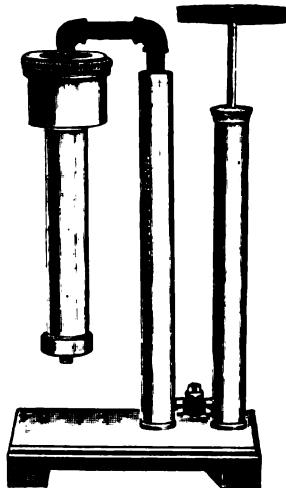
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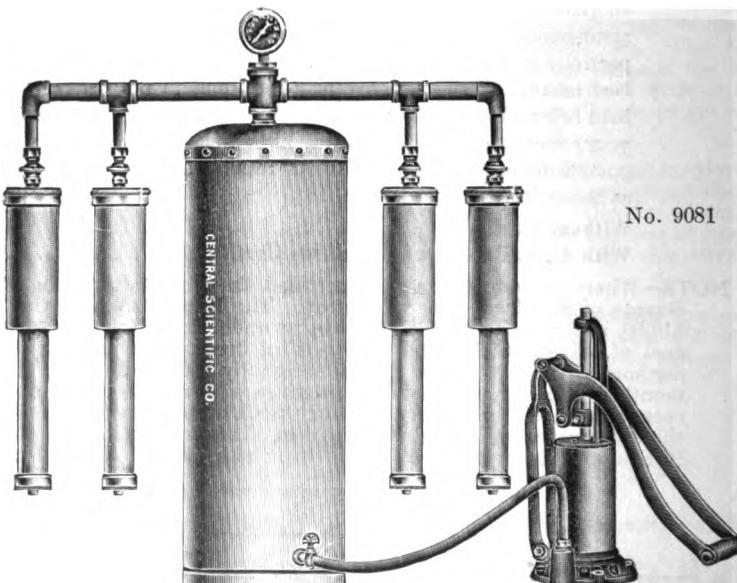
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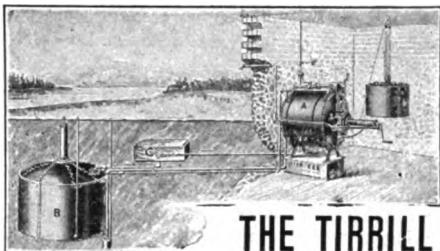
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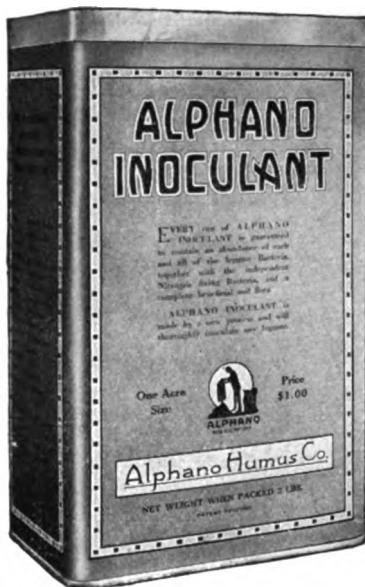
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THE INFLUENCE OF SODIUM NITRATE UPON NITROGEN TRANSFORMATIONS IN SOILS WITH SPECIAL REFERENCE TO ITS AVAILABILITY AND THAT OF OTHER NITROGENOUS MANURES

DAVID A. COLEMAN

From the Department of Soil Chemistry and Bacteriology, Rutgers College¹

Received for publication, May 1, 1917

FOREWORD

For a proper understanding of our subject it is necessary at the outset to realize the conditions and factors influencing plant growth. Plant nutrition is a complicated process, due to the fact that plants synthesize their own plant-food from various substances taken out of the air and soil. These materials consist of carbon dioxide, water, oxygen, and suitable compounds of nitrogen, phosphorus, potassium, sulfur, calcium, iron, and, to a less extent, manganese and silicon.

Aside from the variations in the feeding of the plants, per se, the forces which affect the nutrition of plants may be designated as chemical, physical and biological.

Soils are chemically unlike because of their varied origins and processes of formation. The product of disintegrated rock and decayed vegetable matter, they partake of the nature of the materials from which they are formed. The natural processes of rock disintegration, uneven weathering of rocks of varying-types, combined with the mechanical and chemical action of water, etc., have rearranged, sifted, and sorted the various materials once distributed with some uniformity into soils of many different types. Moreover, not only do they contain mineral plant-foods in sundry combinations, but also air, water and heat. The proper aeration of the soil, its adequate food supply, its warmth, are as essential to plant growth, as is fertility, using the word in the narrower sense. They contribute to its development in that they are among the agencies at work in the preparation of available plant-food. The relationship of soil air in opening up the soil and rendering it permeable to water; the relationship of water to solution, and of heat to ease of solution, are very potent factors in soil fertility. A dense and compact soil being slowly-penetrable by water yields its plant-food slowly and successive crop growth does not occur. Per contra, from a too open soil the water rapidly removes such plant-food as is soluble and the crop starves.

¹ Presented to the faculty of Rutgers College in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

The countless myriads of microorganisms in the soil are important factors in plant nutrition. The modern concept of the soil is that it is a living entity rather than a dead mass, a work shop in a store house, wherein microscopic forms are actively transforming the complex raw materials of the soil into simple forms suited for plant-food. Since their function viewed from the standpoint of soil management is the development of plant nutrients in the soil it follows that soil conditions which favor their growth enhance, and such as retard their multiplication lessen, the crop-producing power of the soil.

The climatic vagaries which are most apt to be harmful are the lack or excess of moisture. Moving through the soil column of varying depths and structure, the amount of this material supplied to plants, as well as the rate at which it is furnished, is of considerable importance to the maintenance of crop yield, regulating as it does the assimilation of food through the roots of the plants.

Of the plant-foods above mentioned, nitrogen is probably the most affected by these physical-chemical-biological forces. Because of the meager supply of this element in the soil and its urgent need for the rational feeding of plants, a knowledge of the influences that these forces exert on nitrogen transformations in soils is of considerable significance.

The economic problem of the nitrogen feeding of plants is of vital importance and becomes more and more urgent as time goes on and populations increase, and their needs become more complex. The physical and chemical influences which regulate the value of nitrogenous manures have been studied in detail by numerous investigators. The biological phase has been touched upon at various times.

It is the purpose of this thesis to consider, under a wide range of conditions, the biological phase of the soil nitrogen problem; noting the effect of fertilizer applications upon nitrogen transformations in soils in the hope that a more proper understanding may be reached with regard to the action of certain fertilizers frequently observed in fertilizer practices.

INTRODUCTION

Nitrogen, the plant-food element of the greatest significance in the nutrition of plants, is fast becoming a limiting factor in crop production. With the increase in the world's population new efforts have been constantly put forth to raise more and larger crops. Consequently, the soil's nitrogen supply is being slowly but surely exhausted and means for its replenishment must assume consideration in the working out of soil problems.

As is well known, there are two main methods by which the nitrogen balance in the soil may be maintained, i.e., the growing of legumes, which when in symbiotic relationship with *Bacillus radicicola* add considerable quantities of nitrogen to the soil, and also the use of nitrogen-carrying materials of different kinds. With the possible exception of the nitrogen which may be

brought down in the snow and rain, and that fixed by non-symbiotic nitrogen-fixing bacteria in soils with proper energy relations, these two above-mentioned agencies are practically the only ones now available in keeping up the nitrogen supply of the soil.

A considerable amount of data has been submitted to show the value of legumes in adding nitrogen to the soil. The experiments, in the main, recommend the growing of legumes more often than is now practised, in order that the nitrogen balance be maintained. Unfortunately, our present economic conditions do not allow us to do this very conveniently, the ordinary three, four and five-year rotation, with a legume once in this period, being the common practice. It would seem, moreover, that the crop immediately succeeding the legume would be the one benefited most advantageously. Indeed, those crops some distance from the legume, in general, are often left nitrogen-hungry.

Necessarily, then, we must look to the second source for supplying us with this most valuable element of plant nutrition. In this country there is spent annually some \$32,000,000 for combined nitrogen. This represents a large outlay of money and well merits the most careful attention of those who are to purchase nitrogen-carrying materials.

In choosing his source of combined nitrogen the purchaser has a wide variety of materials to select from, such as the mineral nitrogen carriers— NaNO_3 , $(\text{NH}_4)_2\text{SO}_4$, CaCN_2 , and $\text{Ca}(\text{NO}_3)_2$ —the ammoniated superphosphates, and the organic materials, of which dried blood, fish scrap, farm and green manures are representative.

From our chemical conception of such materials we would expect rather marked responses from crops to any equivalent quantity of nitrogen in the above-cited materials. Necessarily then, one cannot be too circumspect in choosing the material which would net him the greatest return for the money invested.

PART I

A review of the literature concerning the availability of nitrogenous manures

The earliest fertilizing practices probably go back to the time of the Romans. Kimberley (85) in 1839 writes that saltpetre was known and used as long ago as the time of Virgil. Palissy (134) also remarked as early as 1563, "You will admit that when you bring dung into the field it is to return to the soil something that has been taken away."

About 1650 Glauber (49) in seeking for the principle of vegetation set up the hypothesis that it was saltpetre. Finding this material in the earth cleared from cattle sheds and applying it to soils, he found that it produced enormous yields. He likewise concluded that the fertilizing value of hair, bone, and shoddy was due to the saltpetre it contained.

Kimberley (85) also writes of English farmers testing the effect of different manures as early as 1670, and likewise quotes experiments down to 1828.

During the second quarter of the nineteenth century England and Europe saw the importation of large quantities of guano, nitrate salts and ammonia carriers. The results that these fertilizers produced were very astounding and greater importations took place. English farmers carried out any number of experiments with regard to their value, but like all experiments of this time they showed irregularities hard to explain. Moreover, considerable opposition was experienced from the teachings of Liebig (99), who preached eloquently against their use. Gradually, however, their use became more extant in agricultural practices, thereby bringing to the front the fundamental question concerning their use, i.e., their efficiency, or availability.

The prerequisite for such an understanding was a certain advance in chemical knowledge, which happily took place. Chemical analyses showed that the various nitrogenous manures varied greatly in their nitrogen content, and likewise in their fertilizing value. Tables were therefore constructed with the values of the manures in the order of their nitrogen content. No value was assigned to the character of the nitrogen, it seeming immaterial whether it was in the form of nitrates, bones, leather, rape seed or whatnot.

As experimentation went on, however, it was noticed that manures with equal nitrogen contents had very different fertilizing values. Guano exceeded all others in fertilizing value, and its action was, likewise, more quickly perceptible than that of any of the other forms supplying the same amount of nitrogen; something in the makeup was lacking in the others. Chemical analysis showed it to be rich in ammonia and since ammonia was already known as a plant-food it seemed quite easy to explain the rapid action of guano.

From this time on then, there was a distinction between what Liebig (99) called digestible and undigestible plant-food. The former included ammonia and nitrates, and the latter those which became available only when the nitrogen was converted into ammonia.

The fact that they now had a definite compound which they could weigh and calculate upon was a distinct advance in the availability question, allowing extensive experimental investigation. We thus had an unending number of field experiments in England and upon the Continent, the various nitrogenous materials including NaNO_3 , KNO_3 , $\text{Ca}(\text{NO}_3)_2$, $(\text{NH}_4)_2\text{SO}_4$, $(\text{NH}_4)\text{Cl}$ and a large number of organic nitrogen carriers such as the bloods, fish scrap, rape meal, and the guanos being compared as to their value for the nitrogen feeding of plants.

To these early experiments belong the work of Lawes (96) in England, Boussingault (15) in France, S. W. Johnson (96) Ritthausen, (153) Knep, (88) Deherain, (24) Maerker, (118) and Ville (201). These investigators, testing out the various nitrogenous manures on hundreds of soils, with different crops and under different climatic conditions, came to the conclusion that an equal quantity of nitrogen in the form of nitrates was better than the same quantity of nitrogen as ammonia, and this in turn was better than an equal quantity in organic forms.

However, there is much justifiable objection to this early work. In many cases no provision was made for the elimination of all but one limiting factor. Uniform soil conditions and the presence of an abundant supply of mineral plant-foods, were lacking in many instances, making the results open to question and criticism.

In 1881 Wagner (209) was led to propose the elimination of the cumbersome field trials and substitute for them pot and cylinder experiments. These methods of study, he argued, would allow of the more perfect control of the various factors in crop production. The tenability of his claims was only too apparent and thus we see the development of pot and cylinder experiments on a large scale by investigators the world over.

Wagner (208) himself carried out an immense amount of research on the nitrogen feeding of plants, directing especial attention to the value of nitrate of soda as a nitrogenous manure in comparison with a large number of fertilizing materials. His work entailed a study of the recovery of the nitrogen applied in different forms by numerous crops, among which may be cited rye, barley, oats, winter wheat, mangolds, spring wheat, mustard, carrots, potatoes and sugar beets; and under numerous soil conditions such as moisture, temperature, reaction and physical and chemical composition.

As a general thing he found in round numbers that out of every 100 parts of nitrogen supplied to the crop in the form of nitrate of soda, 70 parts were returned in the crop, whereas, with ammonium sulfate from the same 100 parts applied, he found returned in the crop 56 parts. This recovery, however, varied slightly with his crops; root crops or those crops having a long-growing season giving a higher recovery than short-season crops. His nitrogen returns in fertilizing oats were 74 parts, with rye 61, wheat 60, barley 64, mustard 55, rape 57, flax 77, potatoes 90, and carrots 91, when 100 parts of nitrate of soda were used. Similarly when ammonium sulfate was supplied, out of every 100 parts there was returned in oats 61, rye 56, barley 54, mustard 51, rape 39, flax 64 and carrots 72. His recoveries with the other organic manures were correspondingly smaller, varying with the quality of the materials supplied.

Taking into consideration all of the factors which would be involved in the action of these manures, such as a deficiency of lime in the case of ammonium salts, liability to leaching of nitrates, the gradual action of organic manures, which is quite desirable in some cases, he worked out a table of efficiencies giving the recovery from nitrate of soda a value of 100. Basing his other figures upon this he gives for ammonium sulfate a value of 90, for dried blood a value of 70, manure 45, and wool and leather 30 and 20, respectively. Checking his pot and cylinder experiments against field trials, with the same fertilizers he obtained the same order of availability although his relative values became somewhat lower. In his field trials ammonium sulfate was assigned a value of 75 instead of 90, as was found in his pot work.

As reported in another publication (211) he repeated his first researches

in 34 distinct experiments on 9 different soils and came to practically the same conclusions as he had previously reached.

However, in giving out these values he warns against generalizing too far. Organic nitrogen, he argues, cannot always be substituted for nitrate nitrogen, and conversely, nitrate nitrogen can not always replace organic nitrogen.

A vast amount of research has taken place since Wagner (209) first suggested his principles for studying the values of fertilizers. Some of the more important contributions are summarized in the following pages, particular attention being paid to the investigations of the last twenty years. For the sake of coherency the literature is summarized in groups which take up in turn: (1) the relative ability of $(\text{NH}_4)_2\text{SO}_4$; (2) the value of the new synthetic manures CaCN_2 and $\text{Ca}(\text{NO}_3)_2$; (3) the efficiency of the higher grade organic materials—dried blood, rape meal, cottonseed meal, etc., and (4) the lower grade nitrogen carriers—animal excrement, wool waste and the like.

Following closely upon the publication of Wagner and Dorsch's (208) work in 1892 we find Grandjean and Bertman (52) reporting along similar lines from France. In a field experiment for the seasons 1892–1897, growing corn, oats, wheat and potatoes, they compared the relative values of NaNO_3 , $(\text{NH}_4)_2\text{SO}_4$ and dried blood, and assigned for them the figures 100, 82.90 and 71.30.

A year later, Thorne (192) in a 2-year field experiment compared NaNO_3 and $(\text{NH}_4)_2\text{SO}_4$ on a rotation of corn, oats, wheat and clover and reports a slightly higher value, namely 84, for $(\text{NH}_4)_2\text{SO}_4$.

In 1900, Warington (215) in speaking of the fertilizing experiments at Woburn and Rothamsted said that for cereals $(\text{NH}_4)_2\text{SO}_4$ had a value of 93, for hay 88, for mangels 82 and for turnips 100 when the action of an equivalent quantity of NaNO_3 is valued at 100.

Pfeiffer (141), in the same year, reports as a result of a 3-year pot experiment with oats, mustard, and carrots as the crops, that $(\text{NH}_4)_2\text{SO}_4$ should be valued at 87 if NaNO_3 is to be valued at 100.

Von Feilitzen (35), concludes from seven trials with $(\text{NH}_4)_2\text{SO}_4$ in moor soils that it should have a value of 91. In one trial with barley in a loam soil he got a value of 75, and with rye on a sandy loam he obtained a value of 75 also.

Von Sigmund (204) published the results of his pot-experiments in 1904. Using a poor sandy well provided with lime he secured an availability of 90 for $(\text{NH}_4)_2\text{SO}_4$ when wheat, rape and buckwheat were grown.

Krietschmer (93), reporting coöperative experiments from the Bonn, Bernberg, Halle and Koslin Stations in Germany by the Wagner (209) method, confirms in general the latter's conclusions, although noting some exceptions.

As a result of pot experiments comparing the relative values of sodium nitrate and ammonium sulfate for the oat crop, Steiglich (178) would assign ammonium sulfate a value of 95, Sjollema (173), 95, Hansen (61), 91, Soderbaum (175, 176, 177), 88–95, Schreiber (168), 77 and Siebelien (172), 59.5.

For the wheat crop Vauha (200) reports a value of 96 and Soderbaum (175, 176, 177), 88. Likewise Berry (9), Behrens (6), Kuhnert (95), Liebenberg (98), and Schneidewind (164, 165, 166), all report greater values for sodium nitrate than ammonium sulfate for the growth of small grains.

As a fertilizer for corn and carrots, Schreiber (168) values ammonium sulfate at 77. In limed soils, however, he raises this value to 81.

Reporting the results of five years of experimentation at the Danish Experiment Stations, Hansen (61) concludes that for root crops ammonium sulfate should have a value of 71.

Somewhat higher results, on the other hand, were obtained by VanHock (198), in the Netherlands. This author finds a value of 99.5 for sugar beets, and for grains a value of 97.5. For potatoes $(\text{NH}_4)_2\text{SO}_4$ gave him better results on peat soils, but on the other types of soil NaNO_3 was superior. Vauha (200), on the other hand, found a value of 66 for root crops, while Von Feilitzen (35) gives it a value of 86 for these crops. Grieg (56) and Schmoegeger (162) also found NaNO_3 superior to $(\text{Na}_4)_2\text{SO}_4$ for potatoes. Malpeaux (121) in testing the effect of these two fertilizers on mangels reports the test results from the fertilization with NaNO_3 .

In a series of field experiments at the Maryland Agricultural Experiment Station, Patterson (137) states that $(\text{NH}_4)_2\text{SO}_4$ has not given as good results as NaNO_3 under any conditions. At Rhode Island Wheeler (220) found that the action of $(\text{NH}_4)_2\text{SO}_4$ was greatly accelerated by the presence of lime. This is likewise the experience of the Pennsylvania Station (221). Regarding the influence of lime upon the action of $(\text{NH}_4)_2\text{SO}_4$, Graudeau (51) says that from his work he would give it a value of 80 in unlimed soils, in moderately limed soils 86, but in heavily limed soils a much lower figure, 63, due no doubt to the loss by volatilization.

Perhaps the most authoritative work in this country on the subject of availability has been carried out at the New Jersey Agricultural Experiment Stations. This work was started by Voorhees and Lipman (205) in 1898, and is in operation at the present day.

In a 15-year cylinder experiment, with three 5-year rotations of corn, oats 2 years, wheat and grass, Voorhees and Lipman (205), and very recently Lipman and Blair (107) have found that by giving NaNO_3 a value of 100, $(\text{NH}_4)_2\text{SO}_4$ should be valued at 65. The fact that this experiment has been running for so long a period and has been carried out under such a diversity of conditions should establish it as one of the most authoritative in the world on the availability question.

Others who have contributed to the availability question showing the superiority of NaNO_3 over $(\text{NH}_4)_2\text{SO}_4$ are Bassler (5), Petermann (139), Hasselhof (66, 67), Eckenbrick (30), deGrazia (54), Gaskill (39) and Gerlach (40).

It is not difficult to find experiments where $(\text{NH}_4)_2\text{SO}_4$ has given results equal or even superior to NaNO_3 .

Otto (133) reports that in light sandy soils, in warm wet seasons $(\text{NH}_4)_2\text{SO}_4$

gave him better results than NaNO_3 . Kelley (80), Kellner (84), Treles (194), all report the better action of $(\text{NH}_4)_2\text{SO}_4$ in the growing of rice.

Popisilki's (142) work shows the same general phenomena, as well as that of Kleberger (86), von Feilitzen (35) Kreitschmer (93), namely, that on open sandy soils in climates of abundant rainfall, $(\text{NH}_4)_2\text{SO}_4$ is liable to give better returns than NaNO_3 .

Clausen (18), Wehnert (219), Bachmann (4), Suchting (187) Soderbaum (175, 176, 177), Lilenthal-Gueithin (100), Orchimkov (132) and Wein (216, 217), all report $(\text{NH}_4)_2\text{SO}_4$ equal or superior to NaNO_3 in its action.

These numerous experiments with $(\text{NH}_4)_2\text{SO}_4$ show a wide range of returns, in some cases going above, sometimes below, and in other cases equal to NaNO_3 . We may, with propriety, inquire into the underlying causes of such results.

In attempting to explain such phenomena Wagner (208) considers the presence or absence of lime of the greatest importance. In one of his experiments he found that without lime $(\text{NH}_4)_2\text{SO}_4$ has a value of 28, whereas when lime was present, this same relative efficiency became 90. This same opinion finds credence in the work of Clausen (18), Bassler (5), Deherain (24), Graudeau (51), Hasselhoff (66, 67), Hasselbarth (65), Patterson (137), Wheeler (220), Volecker (203), Thorne (193), White (221), Lipman and Blair (107) and a host of others too numerous to mention. This great necessity of lime as an aid to the rapid nitrification of fertilizing materials has also been shown in the laboratory by Müntz and Girad (129), Schloessing (160), Withers and Fraps (224), Warington (215) and others. A good bibliography concerning the influence of lime on the nitrification process may be found in the work of Kopeloff (91).

A second theory to the effect that the inferiority of $(\text{NH}_4)_2\text{SO}_4$ as compared with NaNO_3 finds support in the teachings of S. W. Johnson (76). This investigator believes that the inferiority of ammonium sulfate is due to its becoming fixed in the clayey portions of the soil. This view is also held by Wagner (211), Pfeiffer (140) and Löhnis (115). Pfeiffer found that, in some cases, the ammonia became so securely fixed, to added zeolites of calcium, that the plants were not able to use it.

This explanation is somewhat at a variance with our knowledge of soil nitrification processes. We know from Dyer's (29) work at the Rothamsted Station that nitrification proceeds very rapidly. In fact this investigator writes "the application of ammonium sulfate is in fact virtually tantamount to our application of nitrates." If such be the case very large amounts cannot be held very firmly fixed in the soil.

Yet another theory advanced by such authors as Schneidewind and Meyer (164), and Gerlach and Vogel (45), is to the effect that certain plants have selective tastes for either nitrate or ammoniacal nitrogen. Again we know from investigations of Godlewski (50), Tacke (190), and Immendorff (74), that in the nitrification process there may be a loss of elementary nitrogen. If such is the case the differences in availability are easily comprehended.

As ammonia and nitrates are quickly transformed into protein forms within the microbial cell, we have yet another explanation for the differences in the fertilizing values of the two salts. Being taken out of the system by the microorganisms, it may or may not become available again during the current growing season. Moreover, the remineralizing of the nitrogen is attendant with the same liability to loss as is ammonium sulfate itself. With regard to this last-mentioned theory Wagner (211), thinks it is of a minor nature. He seems to be supported in his contention by the work of Vogel (202). Contrary to this, however, are a number of experiments to show that soluble forms of nitrogen are readily assimilated when suitable energy relations are present. In this connection one may cite the works of Biereima (10), Doryland (27), Stocklassa (179), Lutz (117), Stutzer and Rothe (186), and Kruger and Schneidewind (94).

Lemmermann (97) and his collaborators have shown that slight losses of ammonia occur in soils treated with heavy applications of lime and ammonium sulfate. They are of the opinion, however, that under practical field conditions this will not happen. Hall and Miller (60) found that more ammonia collected in vessels containing acid when placed over fields recently manured with ammonium sulfate, than on the untreated plots. Ehrenberg (32) records that when large quantities of ammonium sulfate and calcium oxide are applied to soils there are considerable losses by volatilization, varying with the wind. Potter and Snyder (143), in reviewing this literature, state that the results are conflicting and proceed to show that under ordinary field conditions appreciable quantities of ammonia are lost.

Wagner (208), also points out that applications of mineral nitrogenous fertilizers may even entirely fail. It may happen that the peculiar effect of the nitrogen taken up by the plant is restricted to an increase of the total nitrogen of the crop, without at all leading to an increase in crop yield. It is clearly brought out by him that when the soil itself is capable of furnishing enough of nitrogen for practically a maximum plant growth, then the addition of sodium nitrate or any other soluble form of nitrogen cannot materially increase the quantity of the produce.

Summarizing the explanations for the differences in availability between sodium nitrate and ammonium sulfate we have: (a) the lack of lime, (b) fixation by soil zeolites, (c) selective preference for the source of nitrogen, (d) loss of elementary nitrogen in the nitrification process, (e) the transformation of soluble nitrogen into microbial protein, (f) loss of free ammonia in soils heavily limed, (g) lack of effort due to the soil's own nitrogen supply.

The instances where ammonium sulfate has shown to better advantage than equal quantities of nitrogen in sodium nitrate are not hard to explain. We may conceive of conditions which militate against the very advantages of sodium nitrate. Its ready solubility, ease of diffusion, renders it readily subject to removal in periods of *excessive* rainfall. Ammonium salts, on the other hand, because of the fact that the very valuable portion becomes tem-

porarily fixed, is less liable to loss by leaching. Thus on open sandy soils, sodium nitrate has been found to give inferior results compared with ammonium sulfate if the seasons are wet. Again, on heavy soils, continued one-sided fertilization with sodium nitrate may leave residues that are detrimental to the physical structure of the same.

Of comparatively recent origin are the newer synthetic nitrogenous manures, calcium cyanamid and calcium nitrate. Their recent entrance into the fertilizer trade has not militated against an accumulation of experimental data to attest their value. In reviewing this portion of the availability literature, experiments with calcium cyanamide seemed to hold an important place.

Wagner (210), who was among the first to experiment with this new fertilizer from a result of pot experiments with carrots, oats, barley, fodder and sugar beets, on nine different kinds of soils, concludes that it was equal in value to ammonium sulfate having a relative availability of about 90. He advises, however, that it be applied several days before planting and only upon soils in good physical condition.

In direct contradiction to this is the work of Svoboda (189) and Kuhnert (95). Svoboda, testing this manure on the growth of fodder beets and potatoes, obtained absolutely negative results; Kuhnert found in fertilizing sugar beets that he obtained a financial loss. With oats he secured somewhat better results, but the increase did not pay for the cost of the fertilizer. These negative results are no doubt due to the fact that they used large quantities on very poor soils.

Schneidewind (166, 167), Soderbaum (175, 176, 177), Vauha (200), Rhodin (150), Urban (197), Rossler (154), Hansen (61), Libenberg (98), Schulze (170), Sebelien (172) and Sjollema (174), found somewhat lower values with the use of this material. Hansen, Schneidewind, Urban, and Vauha record a value of 41, 68.5, 86 and 58, respectively, when NaNO_3 is considered as equal to 100. Urban, however, finds that his value fluctuates with the rainfall, reaching much lower values in dry seasons.

For the oat crop Soderbaum (175, 176, 177) gives a value of 92, for rye from 44 to 69, and for wheat 61 to 68. Schneidewind (166, 167) gives it a value of 89 and Vauha (200) 89 for wheat. Likewise Stutzer (185) and Sebelein (172) have also reported experiments to attest the value of $\text{Ca}(\text{CN})_2$ on small grains. The former records a value of 82 for the oat crop and the latter 54, all the above values being calculated on NaNO_3 as 100. On the other hand, Hendrick (70) and Molinari (128) report that $\text{Ca}(\text{CN})_2$ was as effective as $(\text{NH}_4)_2\text{SO}_4$. Others who have reported similar conclusions are Müntz and Nottin (130), Uchiyama (196), Rhodin (150), Sjollema and de Weldt (174), and Bassler (5). Sjollema (173) reports to a value of 120 for it with NaNO_3 as 100 and Bassler rated it at 103 from the results of his work.

Soderbaum (175, 176, 177) reports that with small and large amounts of the fertilizer, he obtained inferior results as compared with NaNO_3 , but with intermediate amounts, 50 pounds per acre, he received better results.

As a top-dressing material this fertilizer seems to be very unfavorable. Gilchrist (48), Grieg (56), Schultze (170), and Wein (216) all report injurious effects from its use for this purpose, due to the very drastic action it has upon the foliage.

A survey of the literature also seems to indicate that some little time needs to elapse between the application of this fertilizer and the planting of the crop. Behrens (6), Haselhoff (66), Kahn (78), Knierim (87), Otto (133), Steglich (178), Wagner (210) and Zeilstroff (226) have published work relating to the time which should elapse between application and planting. Behrens thinks that six days are enough, while Haselhoff goes so far as to advise fall application. Kahn would recommend thirty days in light sandy soils and Knierim, Otto, and Zeilstroff recommend from one week to fifteen days.

Soil conditions also seem to be a much more important factor for the best action of this manure than of NaNO_3 or $(\text{NH}_4)_2\text{SO}_4$. Immendorf (75) says it is not adapted to acid or light sandy soils. This idea finds confirmation in the work of Sebelien (172) who gave it a value of 22 for the mustard crop in a light sandy soil. Remy (148) is of the opinion that its most favorable action is on clay soils. He says that four weeks are necessary to overcome its toxic effects. Uchiyama (196) concludes that neutral soils are best and sandy soils are very bad for its action. This is especially so in dry seasons Gilchrist (48) reports, and is supported in this respect by Urban (197) and deGrazia (53).

A summary of the effect of lime nitrogen as reported by various European investigators is to the effect that it is not adapted to acid humus, or light sandy soils. Its action, however, can be improved on such soils by means of lime, the best results being obtained on fine-textured soils, rich in lime and organic matter, and well supplied with microorganisms which can convert it into ammonia and nitrates. As a general average it would seem that it had a fertilizing value somewhat inferior to $(\text{NH}_4)_2\text{SO}_4$, the best results being obtained with potatoes, the poorest with beets, and intermediate results with grains. When bacterial action is deficient dicyanamid if formed which is very injurious to plants. If moisture is a factor or lime is deficient ammonia is formed by chemical means. Thus the need of a potent soil flora is very evident for the success of this fertilizer.

Calcium nitrate in all possibility occupies a position in the availability table similar to that of NaNO_3 . A number of investigators have reported that in soils deficient in lime this fertilizer gives results quite superior to NaNO_3 . This seems to have been the experience of Wagner (211) and Stutzer (184, 185), in their availability studies on potatoes, oats and sugar beets. Schloessing (161), Sebelien (172), Steglich (178), Schneidewind (166, 167), Passerin (136), Limay (101), Stohr (181) and Belleroux (8), all conclude that $\text{Ca}(\text{NO}_3)_2$ is equal to NaNO_3 . There are some dissentors, Greig (56), Rhodin (151), Hansen (61), Dudgeon (28), and Bassler (5) finding that NaNO_3 gave them the best results. Grieg (56) finds that $\text{Ca}(\text{NO}_3)_2$ did not give him good results on potatoes and Rodin (151) notes that on crops like oats, not requiring lime to any great

extent, $\text{Ca}(\text{NO}_3)_2$, was inferior to NaNO_3 . Hansen (61) reports a value of 76 for roots and 88 for grains. Bassler (5) commenting on its use in moor loamy soils for rye and oats, gives it a value of 96 for both crops. Gerlach's (43) field experiments would give it a value of 74, whereas his pot experiments value it at 103. Soderbaum (176, 177), also considers $\text{Ca}(\text{NO}_3)_2$ of less value for grains, giving it a value of about 96. Schneidewind (166, 167) and Rhodin (151) in later papers give it a value of 96 and 98, respectively. It would seem then, taking everything by and large, that this fertilizer is practically equal to Na_2NO_3 , provided its poisonous principles, i.e., nitrites, are absent.

The organic sources of nitrogen are as numerous as the experiments recording them. Under this heading come the effectiveness of the various guanos, blood, bone, fish and meat meals, farm and green manures, as well as the extremely inert substances such as peat, leather and wool waste.

In discussing the availability of the guanos Wagner (208) says that fish guano has a value of 78 when $(\text{NH}_4)_2\text{SO}_4$ is valued at 100. The same author gives Dumara guano a value of 91, and Peruvian guano a value of 87. Schneidewind (166) considers animal guano at 67 and Pfeiffer (141) sees in fish guano a value of 64. Most of these experiments with guano seem to have been carried out with no attention to the fact that this material also contains large quantities of phosphoric acid. In this connection might be cited the work of the Rhode Island Experiment Station (63) on the phosphoric acid value of different guanos.

Various authors have worked upon the efficiency of farm manures. Gerlach (41) gives stable manure a value of 41, and cow manure 18, when NaNO_3 is considered at 100.

Voorhees and Lipman (206) differentiate the values of the different portions of manure, assigning a value of 35.9 for solid fresh manure, for the liquid and solid together 53, and liquid portion leached 43. Lipman and Blair (107) continuing the experiment, have obtained similar figures.

By means of pot experiments Von Sigmond (204) also obtained data for the value of manure in different conditions; calling NaNO_3 100, he gives for fresh manure in the spring a value of 49, the same applied in the fall 48, rotted manure applied in the spring a value of 42 and for rotted manure applied in the fall 58.

Gilchrist (47) used fresh manure, manure kept long enough to be in good applicable condition and old manure stored in a heap for months, and found that that which had not been stored too long gave the best results.

Rudorf (155) gives cow manure a value of 23.3. Maercker (119) who has also worked on the availability of cow manure gives it a much lower value than this. Pfeiffer (141) gave sheep manure a value of 48.2, horse 29, cattle 26, and barnyard manure 19.2 when NaNO_3 was considered as 100.

Schneidewind (163) in field experiments received a recovery of only 8.13 per cent of the nitrogen applied in three years. Pfeiffer (141), on the other hand, in the course of three years obtained a recovery of 48.4 per cent on

the first year with potatoes, 9.8 per cent and 4.3 per cent, respectively, with rye and carrots the second year and only 0.8 per cent the third year. He recovered in all 63.4 per cent in the three years. His recoveries are abnormal however, because of the open sandy soil he employed and an abundant supply of lime and minerals.

A similar experiment is reported by Schulze (171) who gives much lower results than those reported above. In a 4-year experiment his total recovery was 23 per cent; 43 per cent of this was obtained the first year, 27 per cent the second year, 16.63 per cent the third year and 12.6 per cent the fourth year. The crops grown included oats, barley, wheat, rye, flax, potatoes and sugar beets.

Doiarenko (26) also reports the poor utilization of manure nitrogen, as well as Summers (188) in his work on truck crops.

With regard to liquid manure Mer (125) in comparing urine with $(\text{NH}_4)_2\text{SO}_4$ received the greatest profit with small applications. Pfeiffer (141) gives it a value of 49 and Hansen (61), regards its fertilizing value as 65 for roots and 72 for grains.

The peats have been given considerable attention as a fertilizing material. Because of the extensiveness of these reports it seemed advisable to see just what value various investigators ascribed to them. Herman (71), comparing rye straw, manure and peat litter and moor peat against no nitrogen, finds that 100 parts of peat nitrogen are as valuable as 2.6 parts of manure nitrogen. This would bring peat pretty low in the availability scale. Lipman (111) likewise finds peat to have a low availability, giving it a value of 10 when NaNO_3 is 100. Hoc (72) finds peat nitrogen to be valueless other than on heavy soils where it has perhaps a beneficial physical influence. However, some investigators seem to think that peat nitrogen can be made available by composting it with fermenting manures. For references to this practice the author calls attention to the work of Johnson (27) and Weiss (218). Turning our attention to another class of organic manures, the so-called green manures, we note that von Sigmond (204) finds for vetch a value of 78 and for alfalfa 59. Schneidewind (165) reports peas, beans and vetch to have a value of 52.5 and beet-tops 43, when NaNO_3 is taken as 100. Wagner (208, 210, 211) gives green manures a value of 80. The work at the New Jersey Stations (110) values green manures at about 50 to 60, depending upon the crop.

The fourth class of nitrogenous materials consists of dried blood, tankage, cottonseed meal, meat and fish scraps, etc. Wagner (208, 210, 211), considering NaNO_3 as 100, values the nitrogen in blood as 70, that in bone meal and dry ground fish at 60, and that of wool waste and leather at 30 and 20, respectively.

The results of the long-term cylinder experiment at the New Jersey Stations (107) give blood a value considerably below this, 60.7. A yet lower figure is noted by Schneidewind who gives it a value of 56. Von Sigmond (204) in his pot work rates dried blood at 72; likewise Rossler (154), and Hartwell (64) at

Rhode Island values it at 80 per cent of the value of NaNO_3 . Graudeau (51), Thorne (193), Johnson (76), all give it a value between 70 and 80 when NaNO_3 is taken as 100. The other organic manures fall considerably below this and should be considered as nitrogen carriers only under exceptional conditions.

The lesser efficiency of dried blood and the other forms of organic nitrogen as compared with the mineral forms is not difficult to understand. In the complicated process of decay preceding the formation of ammonia there are many conditions under which elementary nitrogen may be formed. The greater the resistance to decay, the more prolonged is the ultimate simplification, and necessarily the attendant chances for loss increase. In so far as the growing crop is concerned, the substances which nitrify more readily will have the nitrogen in a digestible form when the plant has need of it, and not at some time after growth has ceased. This in itself is a big factor in the working out of relative values.

Again the character of the material supplying the nitrogen is of considerable importance in determining its value. Materials of a wide carbon-nitrogen ratio in a fresh condition will give less nitrogen to the growing plant than fresh materials of a narrow carbon-nitrogen ratio, due to the rapid assimilation of nitrogen by soil microorganisms. On the other hand, materials like peat and leather will give the crop very little nitrogen because they are so extremely resistant to decay.

Considering the residual values of the mineral manures, as a whole, we find that they leave very little residue—their action seems to be spent during the season in which they are applied. As proof of this we may consult the work of Hall (58), Lipman (107) and others.

The residual effects of the organic manures may be somewhat greater, due, perhaps, not to the nitrogen supplied but to their influence in improving the physical condition of the soil. Rothamsted reports that barnyard manure applied 35 years ago still gives evidence of benefiting the plant. A close analysis of the conditions, however, shows us that other factors than mere residual effects have entered in.

The lasting effects of manures may not always be desirable. These organic substances which change but slowly necessarily produce soluble material after the crop has been taken off, which may be removed from the land by fall rains and lost to the crop.

While there is, then, a very considerable range of variation in the returns from any given quantity of nitrogen, a more or less definite relation still persists which enables one to classify nitrogenous manures according to the readiness with which they can enter the plant tissue. From what has been related in the text it would seem that there is no longer any doubt as to the superiority of nitrate over ammonia salts, which are in turn superior to $\text{Ca}(\text{CN})_2$ or any of the organic forms.

This entire question of availability is one of extreme importance in all sections where fertilizers are used. Yet, notwithstanding its importance, it

is scarcely appreciated by the average user of commercial fertilizers. To him quantity is the desideratum, with quality considered very seldom. If he would but stop and think of the economic side of the question he would conclude as does Lipman (107) when he says:

According to the schedule of trade values adopted by the New England States and New Jersey in 1915, the nitrogen in ammonia salts is given a value equal to that of nitrate of soda, viz., 16.5 cents per pound, the organic matter in mixed fertilizer is valued at 19 cents per pound, while the nitrogen in fine ground fish, meat, and blood is valued at 23 cents per pound. . . . To state the matter in other terms, if the nitrogen in 100 pounds of nitrate of soda is worth \$2.25 then its equivalent in ammonium sulfate would be worth approximately \$1.78 and its equivalent in dried blood would be worth \$1.53. The trade values however, would make the nitrogen in 100 pounds of nitrate of soda and its equivalent in ammonium sulfate, each worth \$2.25 and the equivalent in dried blood worth \$3.56, the latter being more than twice its value as shown by the results of fifteen years of experimental work. To be sure, under a wide range of field conditions, the result would be less disadvantageous to ammonium salts and certain nitrogen plant-foods.

To the thoughtful person, the question must arise as to whether the present adjustment is a fair one. The plea is made that nitrate nitrogen is lost by leaching, but the results of numerous experiments show that the residual effect of $(\text{NH}_4)_2\text{SO}_4$ is no greater and that of dried blood only slight. Also, the residual effects from farm yard manures are entirely spent after the third year, and if much money is tied up in such fertility it is not an economic success. Even it is granted that on certain sandy soils nitrates are lost in the leaching, still the higher returns would point to the advisability of using it, in smaller, and more frequent, applications.

It will be seen from the above review also that different investigators obtained recoveries from any given fertilizer that were far from constant. Varying proportions of the nitrogen applied being returned in the crops. That is to say, the readiness with which the nitrogen of the applied fertilizer is transformed into plant tissue seems to be influenced by factors which we may call external and internal. The internal factors pertain to the mechanical and chemical composition of the nitrogenous manure and are entirely apart from the character of the medium in which the decomposition takes place. For instance, it was seen that in the same soil and under the same climatic conditions the nitrogen in manure was more available than that in peat, or the nitrogen in blood was more efficient than that of the manure. It need hardly be added here, that the internal factors cannot be controlled as readily as the external factors. However, the latter factors the farmer has some means of controlling. He has it in his power to modify moisture, temperature and aeration conditions sufficiently to hasten decomposition processes and to increase in a large measure the solubility of the nitrogenous materials present in, or added to the soil. The work of Wagner (208, 210, 211), Pfeiffer, (141), von Sigmond (204), Lipman and Blair (107), Voorhees and Lipman (205), has intimated that the reason why more or less of the applied nitrogen is returned to the crop is because of the differences in crops themselves, in their root

systems, in growing season, etc. Wilfarth, Römer and Wimmer (222), on the other hand, would have us believe that the lessened recovery is due to losses entailed in metabolic processes.

Again, the work of Tucker and von Seelhorst (193) has shown us that the moisture supply is very important for maximum recoveries of nitrogen and increased crop growth. In a series of pot experiments where they added 0.5 gm. of nitrate of soda to a soil in combination with the other essential plant-food elements, they found that they got no increase in a crop of oats when only a little water was supplied, the supply of nitrogen in the soil being sufficient for the crop needs, the water and not the nitrogen being the limiting factor. When more water was added the plant made growth and utilized the nitrogen supplied, the added nitrogen, in this case 0.5 gm., increasing the crop 10 grams. One-half gram more of NaNO_3 was then added, but was without effect, the water again being the limiting factor. More water was then applied to the pot containing the first 0.5 gm. of NaNO_3 and the crop yield was increased to 20 gm.; but this does not represent the whole possibility because the application of the second 0.5 gm. of NaNO_3 gave a still further increase of 15.5 gm. Perhaps, then, the moisture conditions in the soils were a factor regulating the amounts of nitrogen recovered by the crop. However, the presence of a too generous supply of moisture may create conditions favorable to denitrification. For a review of the literature and discussions of this phase of the availability question, one is referred to the work of Voorhees (205) and Lipman and Blair (107). With regard to the loss of nitrogen by leaching and its bearing upon the availability question, much has been said and written.

The data of the most wide-spread knowledge were collected by investigators at the Rothamsted Station (57). By means of lysimeters, with which they were able to tap the soil column at any desired depth, they have found what they believe to be an average loss of 30 pounds of nitrogen per annum.

Collison (22) at the Florida Station, however, by means of his lysimeter work, has just recently recorded enormous losses of nitrogen per acre per annum.

McGeorge (123) at the Hawaii Station concludes from his laboratory experiments that the sodium nitrate, not being absorbed by Hawaiian soils, is very liable to be lost in the drainage waters. However, it must be brought to the reader's attention that the conditions of his experiment are so manifestly artificial that their application to actual field conditions is not tenable. The fact that large losses of phosphate and potash salts were found, under the conditions of his experiment, would substantiate this. In his work the possibility of soluble salts rising again to the surface by capillarity were entirely eliminated by his method of studying the problem. That this is an important item can be seen from the experiments of Malpeaux and Lefort (120) who found that quantities of sodium nitrate placed at a distance of 1 meter under the surface of the earth, reached the upper 6 inches of surface soil in less than 6 weeks. This same criticism may apply also to the work of other investigators (22, 57).

Hall (57) likewise states,

On the Rothamsted soil ammonium salts are not retained as such for more than the season of application, nor are the nitrates resulting from them able to return to the surface to feed the succeeding crop. On other soils of better texture for allowing the movement of water by capillarity there can be no doubt that the nitrates in the subsoil will return to the surface and be of service to the crop.

This corroborates the statement of Malpeaux and Lefort (120).

Collison's (22) figures on the enormous loss of nitrates by leaching are open to two criticisms. First, it seems to the writer hardly justifiable to tap the root zone at the point which he did and call the drainage obtained at this point lost to further crop growth. It has been shown by Miller (126) in a study of the root systems of agricultural plants, that many roots reach the point at which these investigators tapped the soil water, and are able to use the plant-food tabulated as lost. Furthermore, the conditions under which the soils were placed in the tanks allow of a very rapid nitrification of the soil organic matter, as has been shown by Deherain (25), and consequently the losses recorded are many times more magnified than would actually be the case.

Hall (57) also seems to show that the movement of nitrates in the ground is not as rapid as is generally supposed; i.e., nitrates in the ground water of treated plots did not mingle with those of the non-treated.

Moreover, it is a question in the writer's mind whether the figures representing losses of nitrates by leaching are truly representative of the facts. If we should consider the total drainage of the rivers of the world (17) we should find that there is annually deposited into the ocean some $24,614 \times 10^3$ metric tons of NO_3 per annum, or $5,513 \times 10^3$ metric tons of the element nitrogen. As this amount of nitrogen comes from a drained area of 40,000,000 square miles, calculated to the acre basis this loss would become very little indeed, i.e., 4 pounds per annum.

However, it cannot be denied that nitrates are susceptible of being driven down to the lower depths of the soil by means of copious rains and by the time they rise again to the surface are of little use to the crop that season. This, of course, would be reflected in availability studies. The question might also be raised as to whether the irregular recoveries obtained by the numerous investigators is due to purely physiological causes or whether it may possibly be effected by bacteriological activities.

A survey of the literature will show us that mineral salts applied to the soil increase to a large extent the already large host of soil microorganisms. We also know from the investigations of Caron (16), Stocklassa and Ernst (180) that the bacterial population under any specific crop is variable. Moreover, the extensive researches of Biereima (10), Hutchinson and Marr (73), Lutz (117), Lipman and Brown (112), Löhnis (114), Ritter (152), Stutzer and Rothe (186) and Vogel 202 have indicated that such substances as NaNO_3 , $\text{Ca}(\text{NO}_3)_2$, $(\text{NH}_4)_2\text{SO}_4$, dried blood, and cottonseed meal serve as a source of food to the hungry hosts of soil organisms, and hence we would naturally ex-

pect a reflection in the growth of the plant. Seeking in nitrogen a portion of their food supply, or even energy for their life processes, they multiply the number of its combinations, compelling it to pass through a series of analytical and synthetical changes. The nitrogen of today, a constituent part of some protein molecule, may tomorrow be part of some simple amino-acid, or may unite with some other nitrogen atom and pass away into the air. Conversely, the simple soluble nitrogen in nitrate or ammonia salts today may be part of a complex protein body in a short while, and so on. There is an ever-changing condition of the nitrogen in soils.

It is because of the intimate relation between soil organisms, soil nitrogen and soil fertility, and the frequently controlling position of soil nitrogen as a factor in crop production, that there is such a strong interdependence of soil microbes and soil fertility. It can be no matter of indifference to us what the effects of these fertilizers are on the several processes of ammonification, nitrification, denitrification, nitrogen assimilation and nitrogen fixation. These processes possess for us a paramount interest because they are concerned with this very element whose fortune is so closely allied to the activities of soil organisms. The discovery and comprehension of the biological forces relating to the application of NaNO_3 to the soil is unquestionably necessary to the solution of many soil problems. An adequate knowledge as to the effect of NaNO_3 alone, and in combination with other fertilizing materials, upon the myriads of soil microorganisms is essential for a clearer understanding of the factors affecting the availability of nitrogenous fertilizers.

EXPERIMENTAL

Experimentation has taught us that higher plants cannot assimilate atmospheric nitrogen or nitrogen in the condition that we find it in the higher protein complexes. In other words, the nitrogen must be in a digestible form before it can be of use in plant nutrition. The agencies by which these protein molecules are broken down are of two kinds, chemical and biological. Whereas the former acts only in a minor capacity, the latter is of the uttermost importance for the simplification of the protein molecule. This process of simplification is carried out by many groups of soil organisms, among the most important of which are the ammonifying group and the nitrifying group. The process by which organic matter is transformed into ammonia we call ammonification. It represents a certain stage in the cleavage process, and because of the fact that the protein molecule must be split to this stage at least before it can be used by the plant, it bears an intimate relation to soil fertility conditions. Moreover, this stage must be reached before further mineralization of the nitrogen can take place. It is of no little concern to us, therefore, how this process of ammonification is influenced by the prevailing fertilizer practices. We know that the presence of a proper supply of nitrogen may be the limiting factor between a rich and a poor harvest.

Because of the fact that NaNO_3 is so extensively used in the present-day fertilizing practices, it is important to become acquainted with the influence that this salt has upon the ammonification processes in the soil. In noting this it should be studied from all possible angles—in acid and alkaline soils, in the presence of phosphates, in potash carriers, and in complete fertilizer mixtures. The influence of this salt upon biological activities at different degrees of moisture and temperature should be studied in detail in order to determine its effect on the various groups and types of microorganisms.

The purpose of the first series of experiments was to study the influence of NaNO_3 upon ammonification. Earlier workers such as Lipman and Brown (112), Temple (191) and Sackett (158) have noted that nitrates increased the ammonifying efficiency of soils. Quite recently Greaves (55) has come to the same conclusion. On the other hand, the work of Peck (138) and that of Crowther (23) seem to indicate a decrease in the accumulation of ammonia due to the presence of nitrates in the soil. Wohltmann (225) and his associates, testing the influence of soils treated with NaNO_3 at the rate of 75 kg. per acre, obtained an increase of over 3 per cent due to this treatment, the Remy (147) method being used. We thus have some inkling as to what to expect from fertilization with NaNO_3 .

SOILS USED

The soils used in this investigation were of seven distinct types. Six mineral soils and one of organic origin were used. The soils represent large areas of land in the various localities from which they were obtained. They were a Carrington loam from the Iowa Experiment Station; a Norfolk very fine sand, and a Norfolk sandy loam from Norfolk, Virginia; a Penn clay loam from New Brunswick, New Jersey; a Sierra sandy loam from Riverside, California; a Wooster silt loam from Wooster, Ohio; and a Muck soil from Great Meadows, New Jersey.

The writer takes this opportunity to thank those who have been so kind as to coöperate in collecting these soils.

The hycroscopic moisture, apparent specific gravity, water-holding capacity, lime requirement by the Veitch Method, nitrate content and ammonia content of the soils used are given in table 1.

In table 2 is to be found the capillary rise of water in the soils in centimeters at the end of various intervals of time. The capillary tubes were 1 inch in diameter and 30 inches in length. The figures in table 2 were obtained by allowing the tubes, after being carefully filled with soil, to stand in 1 inch of water for 42 days. It was thought important, inasmuch as sodium nitrate is a very soluble salt, to note this capillary rise, as a factor in alleviating toxic effects of large quantities of the salt.

TABLE 1

Hydroscopic moisture, apparent specific gravity, water-holding capacity, lime requirement, nitrate content and ammonia content in the soils used

	SOIL NUMBER						
	1	2	3	4	5	6	7
Characteristic.....	Carrington loam	Norfolk sand	Norfolk very fine sand	Penn clay loam	Sierra sandy loam	Wooster silt loam	Muck soil
Hydroscopic moisture (per cent)...	2.35	1.00	0.15	2.28	1.35	1.87	18.00
Apparent specific gravity.....	1.05	1.30	1.52	1.05	1.61	1.25	0.70
Water-holding capacity.....	0.50	0.32	0.34	0.50	0.28	0.42	1.60
Lime requirement...	3,700	2,200	550	1,110	Alkaline	4,300	5,200
Nitrates (mgm. per 100 gm. soil).....	2.66	1.67	Trace	4.78	4.79	2.60	13.30
Ammonia (mgm. of a 100 gm. soil)....	0.77	0.06	Trace	0.53	0.13	0.50	42.17

*Veitch method.

TABLE 2
The capillary rise of water in the soils

SOIL TYPE	HOURS													
	4	36	60	84	108	134	168	241	292	350	518	840	954	1022
Carrington loam.....	16.32	36.19	38.87	42.42	45.21	47.62	50.00	53.95	54.96	59.43	62.84	68.50	69.81	62.81
Norfolk loam	21.59	39.72	45.55	48.89	52.70	55.14	57.58	65.25	64.70	68.58	70.24	72.39	72.89	73.15
Penn loam...	12.95	31.75	36.19	39.72	42.92	45.46	48.00	52.47	54.96	59.94	64.77	70.48	72.13	74.40
Sierra loam...	23.77	49.27	54.96	59.05	62.88	67.94	66.90	74.52	76.81	*	*			
Wooster silt	18.08	40.38	46.99	50.54	53.97	56.88	59.69	63.81	75.21	*				

* Top of cylinder.

METHODS

The methods used in studying the ammonification process were of two kinds; (1) the beaker method as outlined by Lipman and Brown (108), and (2) a modification of the fresh-soil method as proposed by Brown (12). The first method was carried out in following manner:

One hundred-gram portions of the soil were placed in tumblers. Organic matter was weighed out into them and the latter mixed with the soil by means of the shaker devised by the writer and his colleague (103). If non-soluble materials were to be used these were added at the same time as the organic matter and mixed with it. NaNO_3 , however, was added in solution. When the insoluble salts and organic matter were thoroughly mixed in the soil, water was added equivalent to 50 per cent of the water-holding capacity. The tumbler was then covered with a glass plate and incubated for 5 days in the case of sandy

and sandy loams, and 6 days for the other soils. The temperature of incubation was 20 to 22°C. unless otherwise stated. At the end of the incubation period the soil was removed to a copper flask with 250 cc. of water. A piece of paraffin and 5 to 6 gm. of MgO were added and the ammonia distilled off and titrated with N/10 HCl, alizarine sulphonate being used as the indicator.

In the fresh-soil method the difference to be noted was that the air-dry soils were first treated with sodium nitrate for different lengths of time at optimum water contents, and at the end of a given interval organic matter was mixed into the soil as before. The subsequent carrying out of the experiment was the same as previously described.

In order to conform as nearly as possible to field conditions, a series of fertilizer additions had to be worked out which would represent in a reasonably accurate degree the concentrations of fertilizers prevalent in present-day practices. If we could assume that all fertilizer was spread broadcast, we would have had no difficulty in determining the amounts of material to apply. However, somewhat over 85 per cent of the fertilizers are at the present drilled in. As a result we are confronted with a series of concentrations, interdependent upon the width of the layer of fertilizer falling from the drill spout and the number of drilled rows per acre. To illustrate this more plainly let us suppose that we are going to apply 50 pounds of sodium nitrate in planting an acre of corn. An acre would resolve itself into a square of 208 linear feet to the side. Corn is drilled in rows varying from 3 to 5 feet apart. Let us take 3 feet 9 inches as a standard. We would thus have 55 rows of corn per acre. Assuming that the fertilizer is spread to a width of 3 inches and moves to each side 3 inches more and that the total vertical movement is about 9 inches, we would have a block of soil 208 feet long 6 inches wide and 9 inches deep in which the fertilizer acts, or a volume of soil of 78 cubic feet. In a sandy loam having 15 per cent of moisture this volume would resolve itself into 66 cubic feet of actual air-dry soil. Taking an apparent specific gravity of 1.5 this volume of soil would weigh 6130 pounds and 55 such rows would weigh 337,150 pounds. Calculating that one acre of the same soil to a depth of 9 inches would weigh 3,500,000 pounds it is easily seen that our original concentration of 50 pounds has become ten times this, or 500 pounds. By the same method of reasoning smaller or larger applications would result in higher or lower concentrations. For instance, in fertilizing potatoes where 1500 pounds of a 4-8-10 fertilizer are drilled in, or in onion growing where 2000 to 4000 pounds of fertilizer are used, and in other lines of intensive market gardening, the actual concentration may run as high as 5000 to 10,000 pounds per acre. It was decided, therefore, to employ amounts from 50 to 10,000 pounds of sodium nitrate per acre, hoping in this wide range to meet with all concentrations occurring in fertilizer practices.

In calculating the applications 3,500,000 pounds was taken as the weight of an acre nine inches of sandy soils, 2,700,000 for loam soils and 1,750,000 for the muck soil. In all cases except where acid phosphate was employed, chemically pure salts were used.

PART II

Series 1. The influence of sodium nitrate upon the ammonification of dried blood

Table 3 records the data to show the influence of NaNO_3 upon the ammonification of dried blood in seven types of soil. Two per cent of dried blood analyzing 12.48 per cent nitrogen was used as the ammonifiable material. The table has been subdivided into seven sections, each one of which gives the data for the action of sodium nitrate upon the ammonification of dried blood in one type of soil. The fertilizer additions are given in column 2. Duplicate determinations with averages are shown for each soil. In addition, the relative value of the various treatments, based upon a value of 100 for the check, has been calculated.

An examination of table 3 shows one marked difference with regard to the influence of NaNO_3 upon the ability of the soils to ammonify dried blood. The greatest reaction experienced from the addition of sodium nitrate was in the Wooster silt loam. In this soil an application of sodium nitrate equal to 100 pounds per acre increased the ammonifying power of the soil 12 per cent and 300 pounds, 15 per cent. Larger applications, however, did not cause any corresponding increase in the ammonia accumulation. When an amount equal to 5000 pounds per acre was present a decrease in the ammonifying power of 30 per cent is to be noted, whereas twice this concentration depressed the ammonifying power just once again, or 70 per cent. An examination of table 2 shows us that this soil has a very rapid rise of capillary water, and no doubt the toxic effect of sodium nitrate would be of only short duration. Considering the crops grown on this soil and the amounts of fertilizer they generally receive, one need not have any fear that sodium nitrate will influence the ammonifying group in any other way than beneficially.

The soil evidencing the least response to additions of sodium nitrate was the Penn clay loam. A stimulative influence of 2 per cent was experienced with a salt concentration equal to 100 pounds per acre, and this same value held true even with amounts as high as 5000 pounds per acre. As much as 5 tons per acre of the salt did not diminish the activity of this soil's flora to the slightest degree below normal.

The results obtained from the action of sodium nitrate applied to the Norfolk sandy loam soil seem to indicate that sodium nitrate is not an important factor in influencing its ammonifying power with dried blood as the source of organic matter. No marked stimulative action is at hand until the high concentrations equal to 1500 to 2500 and 10,000 pounds per acre are present. With a concentration of 2500 pounds a stimulative influence is to be noted equivalent to 4 per cent above normal. Double this concentration also practically doubled the stimulation. This soil, being a typical early truck soil, must necessarily be heavily fertilized, and the fact that no harmful effects are manifested with such high concentrations is a very happy circumstance.

TABLE 3

Influence of NaNO_3 upon the ammonification of dried blood in the various soils

SOIL POSITION	NaNO_3 PER ACRE	WOOSTER SILT LOAM			PER CENT RELA- TION TO CHECK	NORFOLK SANDY LOAM			PER CENT RELA- TION TO CHECK	PENN CLAY LOAM			PER CENT RELA- TION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
<i>pounds</i>													
1-2	None	98.24	100.50	99.87	100	59.87	64.73	62.30	100	72.09	73.87	72.98	100
3-4	50	103.67	103.56	103.61	103	60.75	63.98	62.37	100	69.42	70.10	69.76	95
5-6	100	115.83	108.70	112.25	112	67.39	67.39	67.39	108	75.65	74.76	75.20	102
7-8	300	114.00	114.00	114.00	115	58.30	62.69	60.45	97	78.00	78.81	78.40	107
9-10	500	112.82	111.39	112.11	112	62.30	63.10	62.70	100	73.33	77.14	75.23	102
11-12	700	112.96	114.11	113.53	113	60.54	65.40	62.97	100	75.12	74.76	74.94	102
13-14	900	113.38	116.11	114.74	115	62.30	65.40	62.97	100	74.76	74.96	74.86	102
15-16	1,200	113.39	112.11	112.75	113	66.58	63.30	64.94	104	75.20	75.37	75.28	102
17-18	1,500	111.83	113.21	112.52	113	64.80	64.80	64.80	104	75.20	75.37	75.28	102
19-20	2,500	112.97	112.87	112.93	113	61.80	55.40	58.60	94	75.71	75.25	75.48	102
21-22	5,000	71.50	70.07	70.78	70	66.70	65.12	65.91	107	77.60	72.62	75.11	102
23-24	10,000	35.32	35.18	35.25	35	66.70	65.12	65.91	107	71.37	75.65	73.51	100

SOIL POSITION	NaNO_3 PER ACRE	SIERRA SANDY LOAM			PER CENT RELATION TO CHECK	CARRINGTON LOAM			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
<i>pounds</i>									
1-2	None	107.36	111.18	109.27	100	136.56	136.00	136.28	100
3-4	50	112.81	111.61	112.21	103	138.71	138.00	138.35	101
5-6	100	109.76	113.57	111.66	102	141.14	140.85	140.99	103
7-8	300	113.67	109.00	111.33	102	143.00	142.30	142.65	104
9-10	500	113.36	114.99	114.18	105	138.66	138.00	138.33	101
11-12	700	113.76	115.76	114.75	105	143.00	142.30	142.67	104
13-14	900	116.08	115.54	115.81	106	142.30	142.30	142.30	104
15-16	1,200	113.90	113.60	113.75	104	142.30	142.30	142.30	104
17-18	1,500	103.98	107.14	105.56	95	136.56	139.14	137.84	101
19-20	2,500	103.98	104.38	104.18	95	131.70	131.90	131.80	96
21-22	5,000	93.08	93.28	93.18	85	131.27	128.70	129.98	95
23-24	10,000	89.92	89.62	89.77	82	119.69	117.78	118.73	87

SOIL POSITION	NaNO_3 PER ACRE	NORFOLK VERY FINE SAND			PER CENT RELATION TO CHECK	MUCK			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
<i>pounds</i>									
1-2	None	38.80	38.90	38.85	100	87.32	86.33	86.82	100
3-4	50	40.87	40.37	40.62	104	87.15	85.79	86.47	100
5-6	100	42.94	43.04	42.95	111	86.33	86.00	86.16	99
7-8	300	40.43	41.74	41.08	108	85.54	85.00	85.27	98
9-10	500					86.15	87.00	86.57	100
11-12	700					84.55	84.55	84.55	97
13-14	900	47.90	48.60	48.75	124	86.33	86.90	86.61	99
15-16	1,200	47.96	47.96	47.96	123	86.33	88.39	87.38	101
17-18	1,500	41.85	39.24	40.54	107	86.00	84.00	85.00	99
19-20	2,500	34.11	34.88	34.49	88	86.33	86.00	86.17	99
21-22	5,000	29.43	30.51	29.97	77	84.83	84.50	84.66	97
23-24	10,000	23.32	25.00	24.16	60	83.63	84.14	83.88	96

The ammonifying flora of the Carrington loam differs somewhat more in its response to applications of sodium nitrate than any of the other six soils. When compared with the Wooster silt loam we experienced a gradual response to additions of the fertilizer up to where a concentration of 300 pounds per acre was at hand. From here on, as in the Wooster silt loam, no enhanced or depressed activity was manifested until a concentration of 1500 pounds per acre was supplied. This concentration depressed the activity of the ammonifiers to about normal. A fertilizer increment of 1000 pounds depressed the action about 4 per cent below normal, whereas seven times this caused a depression in the activity of the ammonifying group of about 13 per cent. The capillary rise of water in this soil is not very rapid, as will be seen in table 2. However, considering the fact that only 13 per cent depression below normal occurs with a concentration of 10,000 pounds, per acre, we may have no fear of any harmful effect of sodium nitrate on such a type of soil. In fact, with small amounts we may expect a rather marked beneficial influence to the ammonifying flora.

The Sierra sandy loam is a soil quite distinct from the other types studied. Coming as it does from a semi-arid region, the concentration of the salts in its soil solution is somewhat larger than in any of the other soils. Consequently when the action of sodium nitrate upon the ammonifying flora of this soil is compared with its action in the same type of soil from humid regions, we find high concentrations of sodium nitrate exhibiting toxic effects in the Sierra sandy loam, whereas the same concentrations under the same moisture conditions, do not cause any decreased ammonia accumulation in the Norfolk sandy loam.

A stimulative action is to be noted in this soil, however, the maximum, 6 per cent, being at hand with a concentration equivalent to 900 pounds per acre.

With the Muck soil no decisive response whatever is to be noted. The ammonifying flora is either non-responsive to nitrate fertilization or else the high absorption power of such a type of soil militates against the presence of the salt in the soil solution and its subsequent action upon the soil micro-organisms. Moreover, in spite of the nature of most mucks, this specific soil has a very high concentration of available nitrates and ammonia, as will be seen by consulting table 1. Their presence would also probably inhibit the action of additional quantities of nitrates.

In all of the previous soils, nitrates in some form or other were present. It was thought that the presence of these might have some influence in regulating the reaction of additional quantities of nitrates. That is to say, if the soil was devoid of nitrates its flora would not be accustomed to their presence and fresh additions of the salt ought to give greater and an earlier reaction upon the ammonifying flora.

In order to test this out, a seventh soil, a Norfolk very fine sand was chosen. This soil had only a trace of nitrates in each 100 grams. It had, however, the biological and chemical characteristics of a true soil. It would seem from an examination of section 6 that the *a priori* assumption above made was true.

Although the duplicate determinations in some cases are not all that are to be desired, due no doubt to losses entailed in a soil of such an open texture with this quantity of ammonifiable material, a greater percentage increase is to be noted as a result of the action of NaNO_3 than in any of the other soils examined. The greatest activity was noted at a concentration of 900 pounds per acre. At this point an increase of 24 per cent above normal was recorded. Although exhibiting the greatest stimulation to the presence of sodium nitrate, this soil also was the most seriously influenced in its decomposition powers by larger quantities of this salt. Even though the same moisture conditions are present as are in the other sandy soils a much greater toxic action occurred to depress the activity below normal.

In all of these tests the action of sodium nitrate seems to work along definite lines. We have at first an enhanced effect followed by an action non-divergent from the maximum and later on, as the concentration increases, a depressed accumulation of ammonia. The question naturally arises as to the cause of such a phenomenon. Is it due to internal conditions, such as the composition of the material in itself, or is it due to external conditions, such as the lack of the mineral elements—phosphoric acid, potash and lime? Also, we might find an answer if we studied the action of moisture, temperature or mechanical composition of the soil upon the ammonification of dried blood as influenced by the presence of nitrate of soda. We may further find substance in the theory that perhaps the rapid multiplication of microorganisms in the process of ammonification unavoidably involves the transformation of soluble ammonia back into protein forms.

The tenability of these ideas will be taken up later in the thesis, it being only too apparent that some controlling factor is operating against a larger accumulation of ammonia when the results of the next series are considered.

Series 2. The influence of nitrate of soda upon the ammonification of cottonseed meal

In this series cottonseed meal was substituted for dried blood. Cottonseed meal is a substance of vegetable origin and in this connection represents the plant residues from which the soil organic matter is mainly derived more closely than dried blood, a substance of animal origin representing insect and animal remains. The experiments in this series were carried out in the same fashion as in series 1. Three per cent of cottonseed meal was used as the source of organic matter. The results are given in table 4.

An examination of table 4 shows us that the ammonification of cottonseed meal is influenced very greatly by the presence of added quantities of nitrate of soda. This seems to be true in all but two of the seven soils, viz., Sierra sandy loam and the Muck soil. In these two soils, while an enhanced effect is present, it is not very large.

In this series the soil whose ammonifying flora is the most affected is the Norfolk sandy loam. In this soil there is a gradual and marked stimulation due

TABLE 4

Showing the influence of NaNO₃ upon the ammonification of cottonseed meal in the various soils

SOIL PORTION	NaNO ₃ PER ACRE	CARRINGTON LOAM			PER CENT RELA- TION TO CHECK	NORFOLK SANDY LOAM			PER CENT RELA- TION TO CHECK	PENN CLAY LOAM			PER CENT RELA- TION TO CHECK
		Mgm. N	Mgm. N	Average mgn. N		Mgm. N	Mgm. N	Average mgn. N		Mgm. N	Mgm. N	Average mgn. N	
<i>pounds</i>													
1-2	None	62.19	60.87	61.53	100	29.32	30.94	30.13	100	77.09	77.77	77.43	100
3-4	50	63.72	63.72	63.72	103	39.36	38.39	38.87	122	73.69	74.37	74.03	96
5-6	100	64.61	63.90	64.25	104	40.08	41.14	40.61	131	76.54	78.32	77.43	100
7-8	300	69.42	66.65	68.03	110	42.70	42.70	42.70	140	75.75	79.31	77.53	100
9-10	500	69.54	77.14	73.34	117	41.30	41.30	41.30	139	81.88	81.88	81.88	105
11-12	700	82.47	84.01	83.24	133	48.60	51.84	50.22	167	81.88	80.10	80.99	104
13-14	900	84.01	87.93	85.79	139	54.17	53.94	54.05	179	83.88	83.88	83.88	108
15-16	1,200	88.64	85.79	87.23	141	59.77	56.05	57.91	192	80.99	80.00	80.49	105
17-18	1,500	80.40		80.40	130	62.20	55.40	58.80	195	86.00	85.14	85.57	110
19-20	2,500	86.15	84.55	85.35	139	60.20	59.10	59.65	198	84.37	89.50	86.93	111
21-22	5,000	85.79	85.79	85.79	139	56.60	57.08	56.84	188	89.00	88.33	88.66	114
23-24	10,000	87.39	89.00	88.19	143	63.13	61.98	62.54	207	88.66	85.44	87.05	112

SOIL PORTION	NaNO ₃ PER ACRE	SIERRA SANDY LOAM			PER CENT RELATION TO CHECK	WOOSTER SILT LOAM			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgn. N		Mgm. N	Mgm. N	Average mgn. N	
<i>pounds</i>									
1-2	None	83.27	83.30	83.28	100	64.99	65.05	64.77	100
3-4	50	82.84	83.64	83.24	100	64.91	63.49	64.20	100
5-6	100	83.54	82.05	82.79	99	67.21	70.07	68.64	106
7-8	300	85.52	85.52	85.52	102	69.92	70.10	70.06	108
9-10	500	85.89	85.80	85.84	102	70.88	71.50	71.19	109
11-12	700	85.89	85.29	85.64	102	71.50	71.50	71.50	110
13-14	900	85.46	85.29	85.37	102	76.97	77.20	77.08	118
15-16	1,200	82.84	84.58	83.71	100	80.00	80.98	80.49	124
17-18	1,500	85.56	85.46	85.51	102	83.16	83.02	83.08	128
19-20	2,500	83.34	84.43	83.88	100	87.23	83.22	85.23	131
21-22	5,000	83.16	82.62	82.89	100	81.22	82.35	81.78	126
23-24	10,000	80.11	79.02	79.58	95	67.77	68.94	68.35	106

SOIL PORTION	NaNO ₃ PER ACRE	NORFOLK VERY FINE SAND			PER CENT RELATION TO CHECK	MUCK			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgn. N		Mgm. N	Mgm. N	Average mgn. N	
<i>pounds</i>									
1-2	None	29.87	30.07	30.02	100	51.62	51.80	51.71	100
3-4	50	32.70	32.90	32.80	109	48.94	48.94	48.94	94
5-6	100	34.40	34.89	34.66	115	54.29	54.65	54.47	105
7-8	300	38.15	38.58	38.36	127	53.75	53.00	53.37	103
9-10'	500	42.83	44.38	43.60	145	53.04	52.58	52.81	102
11-12	700	52.32	52.12	52.22	173	52.69	53.23	52.96	104
13-14	900	56.02	59.40	57.71	191	52.51	52.51	52.51	102
15-16	1,200	52.32	45.99	45.94	153	51.89	54.82	53.35	103
17-18	1,500	45.89	45.99	45.94	157	52.69	52.69	52.69	102
19-20	2,500	47.41	47.20	47.30	137	54.10	54.80	54.45	105
21-22	5,000	40.87	41.96	41.41	92	53.66	54.10	53.88	104
23-24	10,000	27.25	28.15	27.70					

to the presence of sodium nitrate, culminating in an increase of 107 per cent above the untreated soil. With the smaller amount of fertilizer, 100 to 300 pounds per acre, there is an increase in the ammonia accumulation of from 30 to 40 per cent over no treatment. In comparing the results of this experiment with those obtained with dried blood on the same type of soil, a most astounding difference is to be seen. In the last series we did not experience an increase of over 7 per cent above normal with all the concentrations used, whereas in this case even 50 pounds per acre gave us an increased activity of over 20 per cent. It is needless to remark again that in no case was there noted any toxicity to the process of simplification of the protein molecule.

The ammonification of cottonseed meal was also greatly enhanced in the Carrington loam by the addition of NaNO_3 . The responses to successive increments of the salt were smaller in this type of soil than in the previous one however. Whereas, in the above soil a 40 per cent increase was obtained with a concentration of 300 pounds per acre, it required several times this amount to produce the same result in this type of soil. Moreover, the maximum stimulation was only 40 per cent above normal, whereas, it was over two and one-half times this in the Norfolk sandy loam. No toxicity from the application of large amounts was apparent.

The Wooster silt loam also exhibited a greater ability to ammonify cottonseed meal in the presence of varying amounts of sodium nitrate than did dried blood. In this type of soil the maximum enhanced activity was 31 per cent above normal. The amount of sodium nitrate present in this case was 2500 pounds per acre. In this soil no enhanced effect was noted at the lowest concentration of applied salts. The reaction is not very marked until a quantity of salt equivalent to 300 or 500 pounds per acre is present. In this soil there is no toxicity present in any concentration to lower the activity of the ammonifying group below the non-treated portions. There is, however, evidence that if the concentration was increased beyond the maximum amount applied here bad effects would follow. This is indicated by the depression from the maximum, 31 per cent, to 26 per cent and 6 per cent, respectively, where 5000 and 10,000 pounds of salt were applied.

In contradistinction to the effect of sodium nitrate upon the ammonifiability of dried blood in the previous series where the Penn clay loam was used, a rather noticeable enhanced activity occurred, reaching a maximum of 14 per cent above normal with an application of NaNO_3 to the highest concentration. Also, appreciable responses are to be noted in this type of soil with smaller quantities. A comparison of the effect of sodium nitrate upon the ammonifying flora of this soil, with the Carrington loam, Norfolk sandy loam and Wooster silt loam shows us that it is the least affected of the four. The question arises as to the cause of such a phenomenon. Is it a question of soil flora? Are the more aerobic conditions present in the three mentioned soils such as to bring out certain types and groups of soil microorganisms, which are stimulated by applications of sodium nitrate, or has the mechanical condition of the soil

tended to adsorb the soluble nitrates thus keeping them out of the soil solution and affecting the soil organisms? Lipman and his associates have shown that applications of sodium nitrate were much more effective in a Penn loam soil that had been diluted by means of quartz sand than in the undiluted portion.

The Sierra sandy loam, as well as the Muck, although evidencing a small enhanced activity due to the presence of soluble nitrates, could not be considered as being very greatly stimulated by the presence of this salt. In the previous series where dried blood was used as the ammonifiable material the Sierra sandy loam recorded a slightly greater enhanced activity than in this series, some 4 per cent less activity being noted here. A toxic effect is also on hand at the highest concentration, being 5 per cent below the untreated. In the Muck soil a more decided stimulating action has been recorded than was present in series 1. A maximum of 5 per cent was reached with the greatest amount of applied salts. With more rational amounts stimulative actions varying from 2 to 3 per cent are to be seen. On repeating the experiment in this series in a soil devoid of nitrates a somewhat similar condition is met with, as was experienced in the previous series. The maximum enhanced effect, 91 per cent above the non-treated portion, was found at a concentration equal to 900 pounds per acre. Fifty pounds per acre increased the activity of the organisms 9 per cent; 100 pounds 15 per cent and 300 pounds 27 per cent. A depressive action is present when the concentration equaled and passed 1200 pounds per acre. This depression does not fall below the record for the untreated portion, but falls off only from the maximum enhanced effect.

Quite apparent is the difference between the action of sodium nitrate upon the ammonification of cottonseed meal and dried blood in all of the seven types of soil used. The question naturally arises as to why this is so. Our knowledge of the chemical constitution of dried blood tells us that it differs quite considerably from cottonseed meal. In the first place, analyses by the writer showed it to have approximately one-half as much total phosphoric acid and potash as cottonseed meal. Also, analyses for total carbon and nitrogen tell us that their carbon-nitrogen ratio is quite different, cottonseed meal having a much greater quantity of available energy-producing materials, i.e., carbohydrates. The writer has shown (21), as well as others, that when organic materials of different carbon-nitrogen ratios are introduced into soils, considerable changes in the microbial flora are brought out. Again, one can conceive of marked differences in the cleavage products of the two substances. Other things being equal, we should expect a greater acid residue from cottonseed meal than from dried blood. This would of course affect the standing of the various groups of soil organisms, as has been shown by Fellers (37) with soil bacteria and Kopeloff (90) with soil fungi.

Perhaps the action of sodium nitrate upon the ammonification of cottonseed meal would not be so striking if the soil were made alkaline with lime. Indicative of such a thing is the action of cottonseed meal in the Sierra sandy loam, which as will be remembered, is alkaline in reaction. As has been pre-

viously pointed out, perhaps phosphoric acid or potash is important for the action of sodium nitrate in increasing the decay of dried blood. With these thoughts in mind we shall follow the decay of dried blood and cottonseed meal in the presence of both acid phosphate and nitrate of soda, in the presence of available potash, in limed soils and in the presence of available energy.

Series 3. The influence of dextrose upon the ammonification of dried blood

Thinking that perhaps the lack of stimulation in the ammonification of dried blood might be due to a shortage of energy-producing materials an experiment was conducted in which energy was supplied in the form of dextrose. However, in order that we might know what influence dextrose exerted upon the ammonifying efficiency of the various soils a preliminary experiment was first made in which dextrose was added in the absence of nitrate of soda. This was carried out by the same method as before. Dextrose was added in amounts equivalent to 0.1, 0.3, 0.5, 0.7, and 1 per cent.

An examination of the data in table 5, recording the influence of dextrose upon ammonification in the various soils, shows us that there is a considerable difference with regard to the influence of dextrose upon the ammonification of dried blood in the various soils. Although the ultimate action of large quantities of dextrose causes a decreased accumulation of ammonia, as has been previously shown by Doryland (27), Lipman and Brown (112) and Hutchinson and Marr (73), in some soils small amounts of dextrose gave a stimulative action to the ammonifiers, and in others no increase above the untreated portions is to be seen. The stimulative action of dextrose in the Penn clay loam is quite marked, 0.10, 0.30 and 0.50 per cent effecting appreciable increases in ammonia production. One per cent decreased the ammonia accumulation 42 per cent. An increased ammonia accumulation was obtained with 0.10 per cent of dextrose also in the Sierra sandy loam, Norfolk sandy loam and the Muck soil. In these soils as well as in the Carrington loam and Wooster silt loam, quantities in excess of this depressed the accumulation in varying degrees. The greatest depression was found in the Wooster silt loam, followed by the Norfolk sandy loam, Carrington loam, Sierra sandy loam, Penn clay loam and Muck soil. It will also be seen from an examination of the table that there are marked differences with regard to the effects of specific amounts of carbohydrates. In other words, 0.30 per cent, 0.50 per cent or 0.70 per cent in one soil did not produce the same percentage of decrease in the other soils. It is evident then that energy-producing material will influence the ammonification of dried blood to a great extent, large amounts producing the same results, whereas smaller quantities cause other effects, not necessarily in the same direction.

The fact that energy-producing materials lowered the accumulation of ammonia from the simplification of dried blood and approached accumulative conditions present with cottonseed meal induced the writer to ascertain whether or not this factor was of importance in causing the phenomena in series 1.

In testing this out, the same methods as previously used were employed. The first series was carried out with small amounts of dextrose. Two-tenths per cent of dextrose was added to all the tumblers with 2 gm. of dried blood, and thoroughly mixed with the soil. Sodium nitrate was then added as before in amounts from 50 pounds to 10,000 pounds per acre, the moisture being brought up to 0.50 per cent of the water-holding capacity of the soil. The incubation and the analysis were exactly the same as in series 1. The soils used and the data accumulated are recorded in table 6.

TABLE 5
Influence of various amounts of dextrose upon the ammonification of dried blood

SOIL PORTION	DEX-TROSE	CARRINGTON LOAM			PER CENT RELA-TION TO CHECK	NORFOLK SANDY LOAM			PER CENT RELA-TION TO CHECK	PENN CLAY LOAM			PER CENT RELA-TION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
per cent													
1-2	None	114.24	113.90	114.07	100	46.80	46.63	46.71	100	42.50	42.34	42.21	100
3-4	0.10	100.74	103.53	102.13	90	47.85	47.89	47.87	102	49.50	51.33	50.46	119
5-6	0.30	80.74	88.08	88.41	77	45.94	44.90	45.42	97	44.71	45.41	45.06	105
7-8	0.50	74.82	74.82	74.82	65	36.71	36.54	36.63	78	42.45	43.65	43.05	102
9-10	0.70	62.98	66.12	64.50	56	30.97	30.27	30.62	65	38.28	38.79	38.53	91
11-12	1.00	53.90	53.90	53.90	47	21.40	21.40	21.40	45	24.70	24.70	24.70	58

SOIL PORTION	DEX-TROSE	SIERRA SANDY LOAM			PER CENT RELA-TION TO CHECK	WOOSTER SILT LOAM			PER CENT RELA-TION TO CHECK	MUCK SOIL			PER CENT RELA-TION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
per cent													
1-2	None	98.65	95.35	97.00	100	92.22	93.96	93.09	100	94.83	94.85	94.84	100
3-4	0.10	102.13	103.85	102.99	105	90.13	90.13	90.13	96	97.20	95.33	96.26	102
5-6	0.30	91.87	91.87	91.87	94	73.08	73.08	73.08	78	89.76	89.43	89.59	94
7-8	0.50	74.82	77.60	76.21	88	56.02	58.63	57.32	61	88.31	84.73	86.52	91
9-10	0.70	69.60	65.25	67.42	75	52.20	50.80	51.50	55	74.64	68.03	71.33	75
11-12.	1.00	49.94	49.94	49.94	51	31.66	30.30	30.98	30	72.10	71.76	71.93	76

A comparison of this table with table 3 (series 1) shows us some striking differences with regard to the influence of sodium nitrate upon the ammonification of dried blood in the presence of available energy.

It will be remembered that in series 1 the effect of sodium nitrate upon the ammonification of dried blood was of this nature—first, an enhanced effect, followed by a reaction non-divergent from the maximum until a toxic condition is reached. However, if we look at the action of this soil when ammonifying dried blood in the presence of dextrose and sodium nitrate, we find an entirely different phenomenon. In the Wooster silt loam we experienced first a slight enhanced effect, and then not the straight line effect of the previous

series, but a decided lessened accumulation of ammonia. With larger amounts we would expect to see the toxic action again acting, and this is the case. The degree of toxicity is different, however, when compared with that in the previous series.

TABLE 6 .

The influence of NaNO₃ upon the ammonification of dried blood in the presence of dextrose (0.2 per cent)

SOIL POSITION	NaNO ₃ PER ACRE	CARRINGTON LOAM			PER CENT RELA- TION TO CHECK	NORFOLK SANDY LOAM			PER CENT RELA- TION TO CHECK	PENN CLAY LOAM			PER CENT RELA- TION TO CHECK
		Mgm. N	Mgm. N	Average m gm. N		Mgm. N	Mgm. N	Average m gm. N		Mgm. N	Mgm. N	Average m gm. N	
<i>pounds</i>													
1-2	None	92.82	93.14	92.98	100	59.29	58.32	58.80	100	60.41	61.21	60.81	100
3-4	50	92.66	91.36	92.01	100	57.89	57.34	57.61	95	57.70	58.98	58.34	96
5-6	100	95.58	96.86	96.22	103	58.83	58.80	58.81	100	57.67	57.67	57.67	95
7-8	300	93.14	92.98	93.06	100	59.61	59.61	59.61	101	56.05	56.00	56.02	92
9-10	500	96.86	97.36	97.11	104	62.37	57.43	59.85	102	60.41	59.60	59.54	98
11-12	700	96.71	96.55	96.63	103	59.77	59.77	59.77	102	56.00	55.08	55.54	91
13-14	900	96.85	97.61	97.23	104	62.53	66.09	64.31	109	55.56	58.33	56.94	93
15-16	1,200	97.84	96.00	96.92	103	63.10	63.50	63.30	108	54.92	56.54	55.73	90
17-18	1,500	94.60	94.00	94.30	101	59.44	58.80	59.12	100	53.10	52.46	52.78	86
19-20	2,500	90.88	90.08	90.48	97	62.37	—	62.37	106	53.10	52.78	52.94	86
21-22	5,000	88.12	88.60	88.36	90	55.89	50.22	53.09	90	52.65	52.60	52.62	86
23-24	10,000	80.19	75.81	78.00	83	61.50	63.40	62.45	106	52.60	52.60	52.60	86

SOIL POSITION	NaNO ₃ PER ACRE	SIERRA SANDY SOAM			PER CENT RELATION TO CHECK	WOOSTER SILT LOAM			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average m gm. N		Mgm. N	Mgm. N	Average m gm. N	
<i>pounds</i>									
1-2	None	103.29	104.56	103.92	100	92.82	92.82	92.82	100
3-4	50	104.13	104.98	104.55	100	92.84	92.84	92.84	100
5-6	100	109.18	109.66	109.42	105	92.84	92.84	92.84	100
7-8	300	101.06	103.68	102.37	99	99.14	92.84	95.99	103
9-10	500	106.46	107.27	106.86	103	95.41	96.21	95.81	103
11-12	700	104.49	106.27	105.38	102	85.05	85.05	85.05	92
13-14	900	110.48	110.80	110.64	107	85.05	81.48	83.26	89
15-16	1,200	109.20	111.50	110.35	107	82.62	82.62	82.62	88
17-18	1,500	107.92	107.10	107.51	104	81.00	81.00	81.00	87
19-20	2,500	103.68	103.68	103.68	100	81.00	81.00	81.00	87
21-22	5,000	83.93	88.90	88.91	85	71.28	71.08	71.17	75
23-24	10,000	76.24	77.86	77.05	75	46.49	46.97	46.73	50

The effect of dextrose upon the ammonification of dried blood as influenced by sodium nitrate in the Penn loam clay soil is also radically different in this series. Previously we had an almost straight line effect. Now the influencing factors cause a broken line effect, the summation of which is a decided decrease in the ammonia accumulation. The influence of dextrose alone was stimulating, that of sodium nitrate alone was not appreciable in any direction.

The algebraic sum is a depression. The most logical explanation would be that in combination these two materials influenced the multiplication of soil organisms to such an extent that consumption was greater than production of ammonia as the nitrate of soda was increased.

From an examination of the effect of these materials upon the ammonification of dried blood in the Carrington loam, one finds no apparent change from the phenomenon in series 1 until a point is reached where the toxic factor again enters in. Comparing the relation values for the three highest concentrations we find that in the series without dextrose they ran 96, 95 and 87, whereas with dextrose the same concentrations of nitrate induced the values of 97, 90 and 83. It would appear that the consumption-accumulation factor was acting again.

We have about the same condition prevailing in the Sierra sandy loam. No marked difference with regard to the combined action of these materials upon ammonification of dried blood is to be noted until high concentrations of sodium nitrate are present. As in the above soil dextrose again lessens the accumulation of ammonia at the highest concentrations of sodium nitrate.

The influence of the combined action of sodium nitrate and dextrose on the ammonification of dried blood in the Norfolk sandy loam and the Muck soil is not different from that of sodium nitrate alone upon the simplification of this material. It will be remembered that dextrose in itself was stimulating; likewise, sodium nitrate in large quantities. The combined quantities of the two do not show any stimulation to speak of over the untreated portions. Thinking, however, that too little energy had been supplied, the series was repeated with the use of a larger amount, 0.5 per cent. The results were of the same nature as previously described.

Interpreted in terms of soil fertility, this series of experiments seems to indicate that in some soils, having an abundant supply of energy-producing materials, nitrate of soda would so influence biological activities that they would secure a considerable portion of the simplified protein, and the plants would suffer accordingly. Moreover, it seems to indicate very strongly that the lack of stimulation is not due to the absence of carbohydrate as was originally surmised.

Series 4. The influence of sodium nitrate upon the ammonification of dried blood in combination with acid phosphate

It will be remembered that the amount of phosphoric acid in dried blood is about one-half of that present in cottonseed meal. Thinking that nitrate of soda would stimulate the ammonification of dried blood or cottonseed meal to a greater extent if more available phosphorus was present, another series of experiments were set up, adding acid phosphate containing 14 per cent of available phosphoric acid to the soil in amounts from 10 to 250 mgm. The same dried blood and method of study were used throughout this series. A pre-

liminary experiment was first set up to determine the influence of phosphoric acid upon the ammonification of dried blood and cottonseed meal before adding the nitrate of soda, the results of which are given in tables 7 and 8. An

TABLE 7
The influence of acid phosphate upon the ammonification of dried blood

SOIL PORTION	ACID PHOSPHATE per cent	CARRINGTON LOAM			PER CENT RELA-TION TO CHECK	NORFOLK SANDY LOAM			PER CENT RELA-TION TO CHECK	PENN CLAY LOAM			PER CENT RELA-TION TO CHECK
		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N	
1-2	None	107.53	108.57	108.05	100	49.41	49.75	49.63	100	54.45	54.45	54.45	100
3-4	0.010	112.05	116.18	114.12	105	57.24	54.63	55.93	112	57.42	57.93	57.67	106
5-6	0.025	112.57	115.85	114.20	105	58.46	57.82	58.14	117	62.64	61.69	62.17	112
7-8	0.050	115.71	117.62	116.66	108	62.64	63.51	63.07	127	60.90	59.86	60.38	110
9-10	0.100	126.32	123.54	124.93	115	67.86	67.86	67.86	136	63.33	64.03	63.68	117
11-12	0.250	128.76	128.60	128.68	119	86.05	86.05	86.05	179	75.60	72.03	73.81	135

SOIL PORTION	ACID PHOSPHATE per cent	SIERRA SANDY LOAM			PER CENT RELA-TION TO CHECK	WOOSTER SILT LOAM			PER CENT RELA-TION TO CHECK	MUCK SOIL			PER CENT RELA-TION TO CHECK
		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N	
1-2	None	111.88	113.10	112.49	100	95.57	94.79	95.68	100	87.00	87.00	87.00	100
3-4	0.010	112.10	113.44	112.77	100	100.92	102.66	101.79	105	91.07	88.74	89.90	103
5-6	0.025	106.58	108.75	107.66	95	101.61	99.18	100.38	104	90.48	90.48	90.48	104
7-8	0.050	118.32	121.80	120.06	106	104.40	111.36	107.88	112	94.48	96.41	95.94	110
9-10	0.100	124.75	122.67	123.71	108	116.58	113.79	115.18	120	94.78	94.94	94.41	109
11-12	0.250	112.92	113.10	113.01	100	109.96	113.48	111.72	115	102.13	103.35	102.74	118

TABLE 8
The influence of acid phosphate upon the ammonification of cottonseed meal

SOIL PORTION	ACID PHOSPHATE per cent	CARRINGTON LOAM			PER CENT RELA-TION TO CHECK	PENN CLAY LOAM			PER CENT RELA-TION TO CHECK	SIERRA SANDY LOAM			PER CENT RELA-TION TO CHECK
		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N		M gm. N	M gm. N	Average m gm. N	
1-2	None	51.60	51.20	51.40	100	34.40	34.60	34.50	100	72.20	70.10	71.15	100
3-4	0.010	53.34	52.40	52.80	101	35.00	35.00	35.00	100	73.40	70.20	71.80	100
5-6	0.025	54.60	56.60	55.60	104	34.40	34.80	34.60	100	69.00	69.60	69.30	98
7-8	0.050	52.60	51.00	51.80	100	34.40	35.40	34.90	100	76.00	76.00	76.00	107
9-10	0.100	60.00	56.20	58.10	112	34.40	34.80	34.60	100	72.60	77.20	74.90	105
11-12	0.250	61.60	58.40	60.00	114	34.40	34.80	34.60	100	68.00	71.00	69.50	98

examination of the tables shows us that in every case applications of acid phosphate increased the ammonification of dried blood. In the Norfolk sandy loam an increase of over 79 per cent is to be noted due to the highest

amount applied, 25 gm. Likewise in all of the other soils there are increases over no treatment varying from 15 per cent in the Wooster silt loam, to 18 per cent in the Muck, 19 per cent with the Carrington loam, and 35 per cent in the Penn loam. At the same concentration in the Sierra sandy loam a depressed activity from the maximum occurred. It would seem then that perhaps a lack of phosphates might be one of the factors limiting the production of ammonia in series 1. This would appear all the more reasonable when we examine the influence of acid phosphate upon the ammonification of cottonseed meal. The soils used to test the effect of cottonseed meal and acid phosphate were the Sierra sandy loam, the Penn clay loam and the Carrington loam. In these three soils the only one in which the ammonification of cottonseed meal was stimulated to any extent at all was the Carrington loam and this took place only at the higher concentrations. In no case, however, was the reaction as large as that taking place with dried blood and acid phosphate. In the Penn clay loam absolutely no increase over no treatment is to be seen. No toxicity was noted in the Sierra sandy loam at this time, as was the case with dried blood and acid phosphate. Taken by and large, the influence of acid phosphate upon the ammonification of cottonseed meal would tend to strengthen the feeling that lack of phosphates was the reason why sodium nitrate did not influence the degradation of dried blood to a greater extent.

However, in order to test the theory out another series of experiments were made. On account of the fact that acid phosphate did not stimulate the simplification of cottonseed meal, the experiment was confined solely to the influence of sodium nitrate upon the ammonification of dried blood in the presence of phosphates. Three types of soil were used, the Penn clay loam, the Sierra sandy loam and the Muck soil. The amount of acid phosphate giving the highest stimulation in the preliminary experiments was supplied to each tumbler with 2 gm. of the same dried blood. Sodium nitrate was then added in amounts equivalent to 50 to 10,000 pounds on the acre basis. The water content was made up to 50 per cent of the water-holding capacity and the contents of the tumblers incubated at 20 to 22°C. for 5 to 6 days. At the end of that time they were analyzed for ammonia as before described. In the untreated portion the ammonia accumulation in the Penn clay loam was 59.50 mgm.; in the Sierra sandy loam 120.40 mgm. and in the Muck soil 96.57 mgm. No deviation from this figure was to be seen in any of the soils when sodium nitrate was added as a limiting factor. As the data are of a negative character it is not deemed advisable to include a table at this point. It would seem, then, that sodium nitrate is not aided in its action by an abundant supply of phosphates.

Perhaps, however, a smaller amount of phosphates or some other materials, such as potash would aid the action of this fertilizer.

Series 5. The influence of potash upon the ammonification process

In order to determine whether potash in itself is of importance for the ammonifying group, series 5 was instituted. The beaker method was used as before, with dried blood as the ammonifiable material. As potassium chloride is probably the most widely used of all the potash carriers, this salt was used in concentrations from 0.01 to 0.25 per cent. Four types of soil were used; the Sierra sandy loam, the Carrington loam, the Penn clay loam and the Muck soil. The incubation and subsequent analysis were carried out as usual.

TABLE 9
The influence of potassium chloride upon the ammonification of dried blood

SOIL PORTION	KCl per cent	CARRINGTON LOAM			PER CENT RELATION TO CHECK	PENN CLAY LOAM			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
1-2	None	126.54	127.42	126.98	100	59.96	60.19	60.07	100
3-4	0.010	125.85		125.85	99	60.87	57.00	58.93	98
5-6	0.025	126.54	124.93	125.73	99	59.28	57.91	59.09	99
7-8	0.050	126.54	126.60	126.57	100	60.87	61.09	60.98	101
9-10	0.100	121.29	119.70	120.49	94	61.56	61.56	61.56	102
11-12	0.250	118.56	117.42	117.99	93	54.72	51.30	53.01	88

SOIL PORTION	KCl per cent	SIERRA SANDY LOAM			PER CENT RELATION TO CHECK	MUCK SOIL			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
1-2	None	128.30	124.26	128.26	100	100.12	98.49	99.30	100
3-4	0.010	131.55	130.41	130.98	103	103.50	103.94	103.67	104
5-6	0.025	132.24	131.10	131.67	104	98.04	100.87	99.45	100
7-8	0.050	133.86	131.58	132.72	105	105.56	105.12	105.34	105
9-10	0.100	125.85	123.80	124.82	98	103.50	103.28	103.39	104
11-12	0.250	93.98	93.98	93.98	79	99.18	99.85	99.51	100

The effect of KCl upon the ammonification of dried blood, as given in table 9, will be considered first. It will be seen that potassium chloride had little or questionable stimulative effect upon the cleavage of dried blood nitrogen. In the Carrington loam no stimulation at all is to be noted. With amounts present equal to 0.01 and 0.025 per cent a depression is noted in the ammonia accumulation. Likewise, in the Penn loam and Sierra sandy loam toxicity is evident at a concentration of 0.25 per cent. On the other hand, appreciable ammonia increases are to be noted as the effect of this salt in small concentrations in the California and Muck soil. This is to be expected, for the California soil is not as yet in a far enough advanced stage of decomposition to furnish all the potash needed, either for the plants or microorganisms. Muck soils are notoriously poor in total potash. Consequently, a response would

naturally be anticipated from the application of potash to these soils. The responses to potash in these two soils and the lack of any response in the other two soils cannot account for the lack of stimulation in series 1. To state the matter in other terms, we could hardly expect potash to benefit the action of sodium nitrate if it in itself cannot increase the degradation of the protein molecule. Parenthetically, it may be pointed out that KCl is greatly more toxic than equal quantities of sodium nitrates.

The above work was duplicated with cottonseed meal as the source of organic matter. The data are given in table 10. In every soil KCl produced some kind of a reaction upon the ammonifying flora. In the Sierra sandy

TABLE 10
The influence of potassium chloride upon the ammonification of cottonseed meal

SOIL POSITION	KCl	CARRINGTON LOAM			PER CENT RELATION TO CHECK	PENN CLAY LOAM			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
<i>per cent</i>									
1-2	None	74.55	71.13	72.84	100	54.72	54.72	54.72	100
3-4	0.010	67.44	66.57	67.00	92	58.56	60.87	59.71	108
5-6	0.025	75.69	75.69	75.69	103	58.14	58.37	58.25	106
7-8	0.050	81.17	81.39	81.28	111	52.66	51.85	52.25	95
9-10	0.100	81.86	79.11	80.48	107	53.05	51.52	52.28	95
11-12	0.250	86.60	84.81	85.70	115	52.44	52.44	52.44	95
SOIL POSITION	KCl	SIERRA SANDY LOAM			PER CENT RELATION TO CHECK	MUCK SOIL			PER CENT RELATION TO CHECK
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N	
<i>per cent</i>									
1-2	None	86.64	85.50	86.07	100	88.79	88.84	88.61	100
3-4	0.010	84.36		84.36	98	87.69	87.69	87.69	99
5-6	0.025	85.95	82.08	84.01	98	89.14	89.36	89.25	100
7-8	0.050	79.35	80.48	79.91	93	90.90	91.94	91.42	103
9-10	0.100	72.96	72.96	72.96	84	95.76	94.16	94.96	107
11-12	0.250	63.80	63.58	63.69	74	89.60	89.82	89.71	100

loam a decided toxic effect was produced because of the presence of all concentrations of this salt. This is in contradistinction to the effect of KCl upon the ammonification to dried blood. With the other three soils a stimulative influence is to be seen in nearly every case. The soil the most benefited seemed to be the Carrington loam, an increase of over 15 per cent above no treatment being registered at the highest concentration of salt employed. In this experiment no toxicity was noted with this soil.

The Penn loam soil exhibited a stimulated action with small quantities of the salt. Larger quantities, however, depressed the activities of the ammonifying flora.

In the muck soil also a slight enhanced effect is present in concentrations around 0.025 per cent. However, if we remember the large stimulative action that sodium nitrate produced upon the ammonification of cottonseed meal in series 2 it does not appear that potassium could have been much of a factor. Koch (89) also has come to a similar conclusion.

Thinking that perhaps single applications did not permit satisfactory procedure in studying the influence of a material, and that a balancing up of the nutrients was needed to obtain better effects from the fertilization with sodium nitrate, another series of experiments were carried out.

Series 6. The influence of various proportions of sodium nitrate, acid phosphate, and potassium chloride upon the ammonification of dried blood and cottonseed meal

In preparing this series an attempt was made to include as large a number of proportions of the three fertilizing materials as possible. This was brought about by the use of the triangular diagram as used by Schreiner and Skinner (169). It has been shown by these investigators that variations in the proportions of three salts by increments of one-tenth of the total concentration will produce a series of 36 separate ratios. A graphic scheme showing the manner in which this series was handled is given in figure 1.

Each side of an equilateral triangle is divided into seven equal parts and lines drawn through each point in the division so obtained parallel to each of the other two sides of the triangle. Each of the original points and each of the intersecting lines represents a distinct and separate culture. All the cultures on the base line of the triangle have one-tenth of the total fertilizer in the form of sodium nitrate, in the line above two-tenths and so on to the top of the triangle which has eight parts of sodium nitrate. The first culture at the base line of the triangle has one-tenth of its total fertilizer concentration in the form of acid phosphate and the second culture two-tenths and so on to the opposite side of the triangle. Thus the eight cultures at the base of the triangle have from one-tenth to eight-tenths of the total fertilizer applications in the form of acid phosphate. Proceeding from the right to the left side of the triangle the partial concentration of potassium chloride in each culture increases in a similar manner as that followed by acid phosphate passing in the opposite direction. Thus the eight cultures in the first row have from one-tenth to eight-tenths of their total fertilizer concentration in the form of acid phosphate and from eight-tenths to one-tenth in the form of potassium chloride, with sodium nitrate making up the remaining concentration.

Each side of the triangular diagram may then be called an acid phosphate side, a potassium chloride side and a sodium nitrate side, respectively.

In using such a diagram it is necessary to start with some arbitrary concentration; after due deliberation a concentration of one-tenth of one per cent was chosen, this concentration being more often than not reached in fertilizing practices. The method of carrying out this experiment was as follows.

One hundred grams of soil were weighed into tumblers and organic matter weighed into them as the occasion might warrant. The three salts were then weighed into them in accordance with that already described, 36 distinct ratios and a no treatment portion being set up, the effects of the single elements themselves having been previously noted. The salts and organic matter were thoroughly mixed into the soil and moisture added equal to 50 per cent of the water-holding capacity. They were then covered with a glass plate and incubated for 6 days at 20 to 22°C. At the end of this time the cultures were analyzed for ammonia in the usual way. However, in order to overcome the time element, it taking some ten hours to complete an experiment of 74 cultures, a scheme was worked out whereby the cultures were analyzed in a definite out-of-order manner.

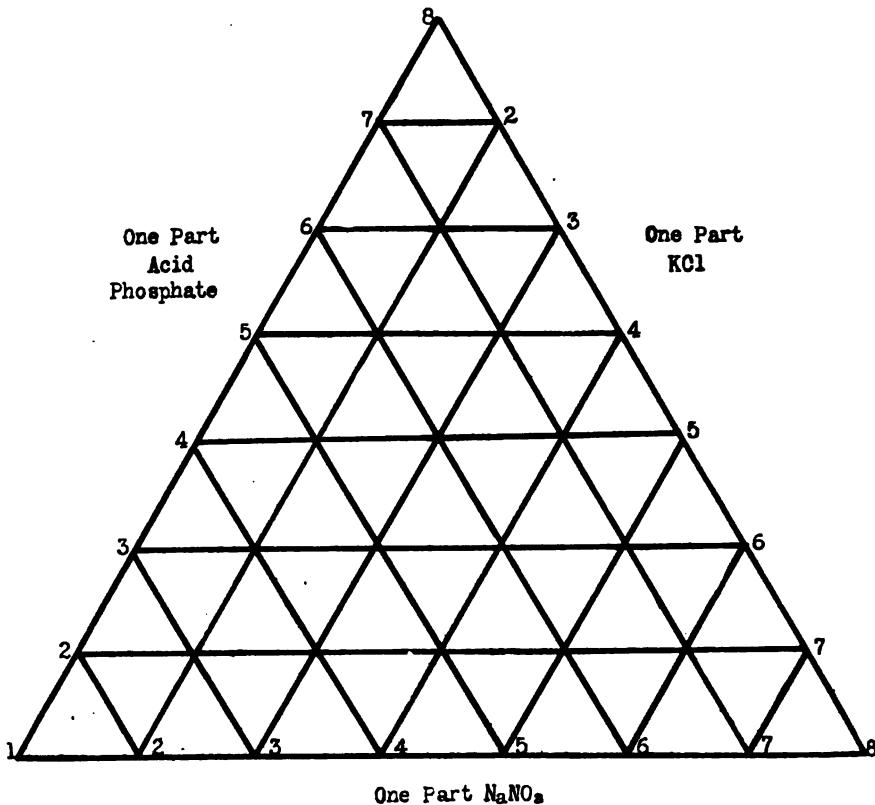


FIG. 1. DIAGRAM SHOWING THE PROPORTIONS OF THE THREE FERTILIZERS, NaNO_3 , ACID PHOSPHATE, AND KCl .

The first experiment was carried out in the Sierra sandy loam with dried blood as the organic matter. The data accruing from this experiment are given in table 11. Column two gives the proportions in numerical values. The values of the specific ratios, the untreated soil being taken as 100 per cent, also are tabulated.

An examination of table 11 shows us that the various proportions of fertilizer carriers all affect the ammonification of dried blood somewhat differently.

Considering first those proportions which give the greatest increases over the untreated soils it will be seen that with an increase in the quantity of acid phosphate in the ratio there is, without any exception, a higher accumulation of ammonia. Comparing also the action of acid phosphate in a complete fertilizer mixture with its action when supplied singly, one finds that the

TABLE 11

The influence of the various proportions of NaNO₃, acid phosphate and KCl upon the ammonification of dried blood in Sierra Sandy loam

SOIL PORTION	PROPORTIONS			ACCUMULATION OF AMMONIA			PER CENT RELATION TO CHECK
	NaNO ₃	Acid phosphate	KCl	Mgm. N	Mgm. N	Average mgm. N	
73-74				110.80	109.44	110.12	100
1-2	1	1	8	105.00	107.52	106.26	96
3-4	1	2	7	111.30	112.56	112.93	102
5-6	1	3	6	121.80	120.66	121.33	110
7-8	1	4	5	127.12	123.90	125.51	113
9-10	1	5	4	126.12		126.12	114
11-12	1	6	3	131.18	123.48	127.33	115
13-14	1	7	2	133.80	135.80	134.80	122
15-16	1	8	1	140.00	138.60	139.30	125
17-18	2	1	7	105.56	113.40	109.48	99
19-20	2	2	6	116.90	119.00	117.95	107
21-22	2	3	5	117.60	117.60	117.60	107
23-24	2	4	4	125.58	125.58	125.58	113
25-26	2	5	3	123.20	127.96	125.53	113
27-28	2	6	2	131.88	131.88	131.88	119
29-30	2	7	1	136.88		136.88	124
31-32	3	1	6	107.64	112.98	110.31	100
33-34	3	2	5	114.52	112.00	113.26	102
35-36	3	3	4	116.48	114.80	115.64	105
37-38	3	4	3	125.30	127.10	126.20	114
39-40	3	5	2	124.88	124.00	124.40	113
41-42	3	6	1	128.80	129.08	128.94	117
43-44	4	1	5	113.54	112.28	112.91	102
45-46	4	2	4	119.00	120.96	119.88	108
47-48	4	3	3	117.60	119.98	118.79	107
49-50	4	4	2	129.22	123.62	121.42	110
51-52	4	5	1	120.96	124.08	122.52	111
53-54	5	1	4	113.88	105.28	109.58	99
55-56	5	2	3	119.00	116.90	117.90	107
57-58	5	3	2	117.60	117.88	117.74	107
59-60	5	4	1	119.56	120.68	120.12	109
61-62	6	1	3	111.44	111.44	111.44	101
63-64	6	2	2	117.12	117.54	117.33	107
65-66	6	3	1	119.84	121.10	120.47	109
67-68	7	1	2	112.28	112.28	112.28	101
69-70	7	2	1	112.28	115.50	113.89	103
71-72	8	1	1	117.88	116.76	117.32	107

presence of sodium nitrate and potassium chloride has greatly benefited the action of this fertilizing material. In the previous series where acid phosphate alone was added to the soil in testing out its effect upon the ammonification of dried blood, it was found that a concentration of 0.10 per cent increased the simplification of the organic matter some 6 per cent. However, the same amount of fertilizer having one-half the concentration in the form of acid phosphate and the remainder in the form of sodium nitrate and potassium chloride, in various proportions, enhanced the degradation of the protein molecule some 15 per cent above the untreated soil. Even in the presence of 0.03 per cent of acid phosphate, which when applied alone stimulated activity only slightly, and 0.07 per cent of various proportions of sodium nitrate and potassium, a 10 per cent greater activity is to be noted. Applied singly, acid phosphate, at a concentration of 0.1 per cent, stimulated the ammonification of dried blood some 8 per cent; with the same concentration of acid phosphate and one part, respectively, of sodium nitrate and potassium chloride, an enhanced effect of three-fold this was experienced, although these last two materials gave very little reaction when applied singly. It will be seen that potassium chloride is toxic at the higher concentrations, which is in keeping with its action when supplied alone.

If we take the proportions where potassium chloride remains constant and acid phosphate and sodium nitrate increase and decrease simultaneously as the total concentration is approached, we find with a decrease in the acid phosphate, a corresponding decrease in the ammonia accumulation. Thus, with a ratio of 1-8-1 an enhanced activity of 25 per cent above the untreated portion occurs, whereas, when the ratio reaches 8-1-1 an activity of only 7 per cent is to be seen; with a gradual decrease between these extremes, showing without a doubt that acid phosphate is the most important material in the mixture.

In this experiment the greatest enhanced activity was manifested with a proportion of 1-8-1 and the least with 1-1-8.

Duplicating the above experiment in the Wooster silt loam, we find a similar set of conditions. The highest stimulation was again found in the 1-8-1 ratio. Making the amount of acid phosphate a constant and increasing and decreasing the sodium nitrate and potassium chloride, we note that at high concentrations of potassium chloride a toxic condition is produced again. Increasing the amounts of acid phosphate seems to stimulate the ammonification of the dried blood and the presence of both sodium nitrate and potassium chloride enhances the action of acid phosphate.

In both of these experiments neither sodium nitrate nor potassium chloride seems to increase greatly the ammonification of dried blood when present in any proportion other than in toxic quantities. Even acid phosphate added in small amounts to these proportions fails to enhance the process. On the other hand, however, acid phosphate seems to create a much greater ammonifying flora when its action is balanced by various quantities of sodium nitrate and potassium chloride.

Another experiment was run, cottonseed meal being used as the ammonifiable material. The results are not suitable for tabulation. It may be said in passing that they strongly indicate that acid phosphate and potassium chloride are of little importance for the ammonification of this material. Sodium nitrate increased the ammonification of cottonseed meal in a manner similar to its action when applied singly.

TABLE 12

The influence of various proportions of NaNO₃, acid phosphate and KCl upon ammonification of dried blood in Wooster silt loam

SOIL PORTION	PROPORTION OF SALTS			AMMONIA ACCUMULATION			PER CENT RELATION TO CHECK
	NaNO ₃	Acid phosphate	KCl	Mgm. N	Mgm. N	Average mgm. N	
73-74				86.80	86.80	86.80	100
1-2	1	1	8	84.00	85.12	84.56	97
3-4	1	2	7	86.52	86.24	86.38	100
5-6	1	3	6	86.94	87.50	87.22	101
7-8	1	4	5	91.98	86.94	89.46	103
9-10	1	5	4	101.08	100.24	100.16	115
11-12	1	6	3	102.79		102.79	117
13-14	1	7	2	105.60	96.88	101.24	115
15-16	1	8	1	105.60	102.77	104.18	120
17-18	2	1	7	86.80	82.46	84.63	97
19-20	2	2	6	88.20	87.78	87.99	102
21-22	2	3	5	91.14	92.12	91.63	105
23-24	2	4	4	91.00	89.74	90.37	104
25-26	2	5	3	93.80	95.48	94.64	109
27-28	2	6	2	95.34	96.60	95.97	110
29-30	2	7	1	105.00	101.92	103.46	119
31-32	3	1	6	88.48		88.48	101
33-34	3	2	5	89.60	89.88	89.73	103
35-36	3	3	4	88.48	92.69	90.53	104
37-38	3	4	3	88.20	90.58	89.39	101
39-40	3	5	2	98.08		98.08	113
41-42	3	6	1	102.84	106.10	104.47	120
43-44	4	1	5	75.78	75.00	75.39	86
45-46	4	2	4	85.82		85.82	99
47-48	4	3	3	84.28	84.28	84.28	97
49-50	4	4	2	90.16	92.97	91.55	105
51-52	4	5	1	95.20	96.74	95.97	110
53-54	5	1	4	82.88	82.88	82.88	95
55-56	5	2	3	86.20	88.20	87.00	101
57-58	5	3	2	94.78	97.30	96.04	110
59-60	5	4	1	93.24		93.24	107
61-62	6	1	3	89.05	86.38	87.71	100
63-64	6	2	2	89.32	87.64	88.48	104
65-66	6	3	1	89.88	90.30	90.09	104
67-68	7	1	2	84.00	84.00	84.00	96
69-70	7	2	1	88.48	88.48	88.00	102
71-72	8	1	1	87.22		87.22	101

Series 7. The influence of alkaline soil conditions upon the action of sodium nitrate in the ammonification of dried blood and cottonseed meal

Perhaps the action of sodium nitrate upon the ammonification process might be different under alkaline conditions. To test this out another series of experiments were set up, the beaker method being used as previously described. In making the soil alkaline enough calcium oxide (C. P.) was added to the soil to neutralize the acidity and leave an excess present equal to 2000 pounds per acre. Although the effect of sodium nitrate in alkaline soil conditions was studied on four soil types, viz., the Carrington loam, Norfolk sandy loam, Penn clay loam and Wooster silt loam, only its effect on the two most acid soils are given.

It will be seen from an examination of tables 13 and 13a that, although lime increased the activity of the soil microorganisms, it did not create conditions favorable for nitrate of soda to stimulate the ammonification of organic matter in either of the two soils. In fact, alkaline conditions seem to diminish the stimulating influence that sodium nitrate exhibits in the same soil, and with the same organic matter in an acid condition.

Moreover, what is very striking is the effect that an alkaline condition induces with regard to the ammonification of cottonseed meal, as influenced by various quantities of sodium nitrate.

It will be remembered that in series 2, the ammonification of cottonseed meal was very largely enhanced by the presence of NaNO_3 . In this series, however, the influence of sodium nitrate is very meager indeed, manifesting itself only at the highest concentrations, and then only to a very slight degree. This fact is also very evident when we consult the data recording the action of sodium nitrate in the limed Carrington loam. In series 2, small amounts of sodium nitrate gave us as great an activity as ten times this amount in this experiment. In the former series the highest stimulated activity was 43 per cent over that of the untreated soil. In this series the greatest enhanced effect is not over 10 per cent above the untreated.

The toxic influence of sodium nitrate is not quite so marked in the presence of lime as can be seen by comparing the respective ammonia accumulations at the highest concentrations in both series. The effect of lime in overcoming the toxic effect of single salt applications has been previously shown by Kearney and Cameron (29), Hansteen (62), Loeb (113), and Oesterhout (131).

As an explanation of the phenomena in this series we may advance several theories. In the first place we may conceive of the combined action of sodium nitrate and calcium oxide so increasing cell division that although there was a marked degradation of the protein the ultimate accumulation would be lowered, because of the fact that it might be used as a source of food and energy.

We know also from the work of various investigators such as Faelli (34), Hall (59), Marchal (123) and Ramann (145), that acid soils may have an en-

tirely different flora from that of alkaline soils. These investigators tell us that acid soils have a much greater proportion of fungi to bacteria than alkaline soils. Brown (11), Engberding (33) and von Feilitzen (36) have taught us that

TABLE 13

The influence of sodium nitrate upon the ammonification of dried blood in limed soils

SOIL PORTION	POUNDS PER ACRE NaNO ₃	WOOSTER SILT LOAM			PER CENT RELATION TO CHECK	CARRINGTON LOAM			PER CENT RELATION TO CHECK		
		Ammonia Accumulation				Ammonia Accumulation					
		Mgm. N	Mgm. N	Average Mgm. N		Mgm. N	Mgm. N	Average Mgm. N			
1-2	None	82.45	81.90	82.17		139.91	138.89	139.40			
	None										
	+										
3-4	lime	104.58	105.28	104.93	100	158.10	158.60	158.35	100		
5-6	100	100.98	103.19	102.08	98	158.10	159.50	158.80	100		
7-8	300	104.21	101.66	102.93	98	161.16	159.80	160.48	101		
9-10	500	106.59	106.27	106.43	102	159.80		159.80	100		
11-12	900	106.23	106.23	106.23	102	154.70	153.30	154.00	97		
13-14	1,500	109.33	107.60	108.46	104	159.90	160.82	160.36	101		
15-16	2,500	93.90	95.88	94.89	89	158.10	156.74	157.42	99		
17-18	5,000	87.04	85.00	81.02	77	153.17	153.34	153.25	97		
19-20	10,000	71.06	69.70	70.38	66	144.88	143.82	144.35	91		

TABLE 13a

The influence of sodium nitrate upon the ammonification of cottonseed meal in limed soils

SOIL PORTION	POUNDS PER ACRE NaNO ₃	CARRINGTON LOAM			PER CENT RELATION TO CHECK	WOOSTER SILT LOAM			PER CENT RELATION TO CHECK		
		Ammonia Accumulation				Ammonia Accumulation					
		Mgm. N	Mgm. N	Average Mgm. N		Mgm. N	Mgm. N	Average Mgm. N			
1-2	None	78.54	83.30	80.92		61.54	57.46	59.50			
3-4	None										
	+										
	lime	89.44	90.93	90.18	100	69.87		69.87	100		
5-6	100	89.44	90.93	90.18	100	69.87	69.87	69.87	100		
7-8	300	93.50	93.50	93.50	103	68.85	68.00	68.42	99		
9-10	500	90.10	92.48	91.29	101	70.55	69.87	70.21	101		
11-12	900	96.22	96.56	96.39	106	67.15	69.36	68.25	95		
13-14	1,500	103.02	101.46	102.24	113	77.86	79.56	78.71	112		
15-16	2,500	96.39	95.20	96.29	106	76.16	73.44	74.80	108		
17-18	5,000	96.56	93.16	94.86	104	74.80	76.50	75.65	109		
19-20	10,000	95.52	95.20	96.36	105	72.76	72.76	72.76	106		

lime changes the flora by considerably increasing the numbers of microorganisms. Moreover the researches of Kopeloff (90), McLean and Wilson (124), Waksman and Cook (212), as well as the writer (21), have shown conclusively that

the addition of vegetable organic matter of various kinds markedly affects the soil flora. It was shown that cottonseed meal and like carbohydrate-carrying materials increase and alter both types and numbers to a great extent. Fungi in particular are greatly enhanced in their activity.

Perhaps, then, the combination of an acid soil and cottonseed meal has produced a flora mainly of a fungus character, stimulated in its activity by application of sodium nitrate. On the other hand, it is known that dried blood is more acceptable to bacterial activity in such a way that we can detect it by this method, or possibly not at all. If, now, we should make the soil alkaline, the activity of the fungi would be depressed and a higher proportion of bacteria would be present. Were sodium nitrate now applied one would expect a reaction similar to that in series 1, if our hypothesis is tenable. Such a phenomenon does actually take place with cottonseed meal, dried blood, calcium oxide and sodium nitrate, particularly with the former source of organic matter. It would seem that the decreased stimulation in this series could be partially accounted for on this basis.

However, in order to gain a more definite knowledge of the action of sodium nitrate upon soil microorganisms, *per se*, another series of experiments were set up to test the effect of sodium nitrate upon some pure cultures of soil microorganisms.

Series 8. The effect of sodium nitrate upon the activity of some pure cultures of soil microorganisms

In studying the influence of sodium nitrate upon the activities of pure cultures of soil organisms the method as outlined by the author (20) in another publication was employed. Briefly, it consists of inoculating sterilized soils supplied with organic matter, with pure cultures of the organisms to be tested and measuring their activity as usual by means of the ammonia accumulation.

In this series cottonseed meal was used, as the stimulation was previously obtained with this material. Ten soil fungi and six soil bacteria were tested out. The *Actinomyces*, being very weak or non-ammonifying organisms, were eliminated. [Consult Waksman and Curtis (213).]

The data showing the influence of sodium nitrate upon the ammonifying efficiency of the soil fungi tested are given in table 14. The organic matter used was cottonseed meal and the soil the Norfolk sandy loam. Sodium nitrate was supplied in amounts ranging from 10 to 250 mgm. per 100 gm. of soil.

It will be seen from an examination of table 14 that soil fungi vary considerably with regard to their response to sodium nitrate. The organism showing the greatest response to applications of this salt is *Aspergillus niger*, an application of 500 mgm. of the fertilizer stimulating its activity 64 per cent over the untreated portion. *Aspergillus bobili*, *Fusarium bullatum* and *Penicillium notatum* were all very favorably influenced by quantities of the salt up to 250 mgm. All the fungi with the exception of *Aspergillus niger* and *Aspergillus*

TABLE 14
The influence of sodium nitrate upon the ammonifying efficiency of certain fungi

SOIL PORTION		ANGERILLUS NIGER		ASPERGILLUS SOIUS		FUSARIUM BULLATUM		PENCILLIUM LAVRATUM		MONILIA SITOPHILA			
		NaNO ₃	Mg ₂ Si ₂ N ₂										
1-2	per cent	None	23.64	23.00	23.32	100	31.10	30.20	30.65	100	63.08	63.08	
3-4	0.010	22.89	22.19	22.54	97	33.67	32.90	33.28	108	62.20	62.40	62.30	
5-6	0.025	24.29	23.80	24.04	104	35.30	36.70	36.00	116	69.42	69.40	69.41	
7-8	0.050	26.26	30.15	28.21	120	36.60	36.80	36.70	117	73.70	73.71	73.70	
9-10	0.100	30.36	30.00	30.18	133	35.89	35.89	35.89	116	73.40	73.80	73.60	
11-12	0.250	33.50	35.40	34.45	149	34.33	34.33	34.33	111	72.50	72.80	72.65	
13-14	0.500	39.69	37.20	38.44	164	39.68	39.65	39.65	127	51.67	51.60	51.63	
SOIL PORTION		MUCOR SPINOSUS		MUCOR SP.		PENCILLIUM NOTATUM		RHIZOPUS TRAITICA		ZYGOMYCHUS			
		NaNO ₃	Mg ₂ Si ₂ N ₂										
1-2	per cent	None	36.32	35.50	35.91	100	41.60	40.50	41.05	100	66.60	64.54	65.57
3-4	0.010	36.25	36.25	36.25	100	43.25	44.65	43.95	107	67.00	69.60	68.30	
5-6	0.025	36.25	35.50	35.91	100	46.89	46.89	46.89	114	70.60	71.60	71.10	
7-8	0.050	37.40	36.00	36.70	101	45.35	45.65	45.50	112	69.00	70.00	69.50	
9-10	0.100	38.40	38.40	38.40	107	45.35	48.90	47.12	114	72.60	73.30	72.95	
11-12	0.250	37.20	37.50	37.35	106	42.10	44.10	43.10	107	72.60	70.00	71.80	
13-14	0.500	31.33	31.33	31.33	89	39.52	39.10	39.31	95	72.60	70.00	71.80	
SOIL PORTION		AFTERAGE, N		PER CENT RELATION		AVERAGE, N		PER CENT RELATION		AFTERAGE, N			
		Mg ₂ Si ₂ N ₂											
1-2	per cent	None	36.32	35.50	35.91	100	41.60	40.50	41.05	100	49.70	49.40	49.55
3-4	0.010	36.25	36.25	36.25	100	43.25	44.65	43.95	104	48.77	48.70	48.73	
5-6	0.025	36.25	35.50	35.91	100	46.89	46.89	46.89	108	49.46	49.46	49.46	
7-8	0.050	37.40	36.00	36.70	101	45.35	45.65	45.50	105	49.46	49.46	49.46	
9-10	0.100	38.40	38.40	38.40	107	45.35	48.90	47.12	111	50.25	50.25	50.25	
11-12	0.250	37.20	37.50	37.35	106	42.10	44.10	43.10	107	72.60	70.00	71.80	
13-14	0.500	31.33	31.33	31.33	89	39.52	39.10	39.31	95	72.60	70.00	71.80	

bobili were affected adversely by additions of 0.5 per cent of sodium nitrate. *Penicillium lividum*, *Monilia sitophila*, *Mucor spinosis*, *Penicillium sp.*, *Rhizopus tritica* and *Zygorhynchus Vuilleminii* were either benefited only slightly or not at all by applications of the salt. None of the organisms, however, gave anywhere nearly as much response to sodium nitrate as was evidenced in the crude culture work.

Turning our attention to the action of sodium nitrate upon the activities of some common soil bacteria we also find a stimulating effect in nearly every case. The greatest stimulation is to be seen in the experiment with *Bacillus subtilis*, and the least with *Bacillus megaterium*. In part this latter organism's activity is greatly decreased by application of sodium nitrate. A fact of importance to be noted is that sodium nitrate is much more harmful to soil bacteria in higher concentrations than to soil fungi.

Considering both groups of organisms, the fungi seem to be more favorably influenced by applications of sodium nitrate than the soil bacteria studied, a fact which would strengthen our hypothesis put forth in the last series of experiments.

In order to see just how far the accumulation-consumption theory is tenable, a new series of experiments were instituted as described in the following paragraphs.

Series 9. A study of ammonia accumulation versus protein simplification as a factor in ammonification studies

In ammonification studies we use as our standard of measure the accumulation of a material readily used by microbial life. Necessarily the amount of material accumulating depends almost entirely upon the excess remaining after the microorganisms have become satiated. In fact, it has been shown, that if the energy relations are right, we may get no accumulation of ammonia at all. This may be the case in the previous series where sodium nitrate did not increase the ammonia accumulation after a certain maximum has been reached.

If we wish to eliminate this factor from our experiments, we must chose some other end point, one which will not be generally used by soil micro-organisms. The bacteriologist has several such processes at his command. He may use the accumulation of carbon dioxide as proposed by Van Suchtelen (199) and Potter and Snyder (143), or he may use the accumulation of sulfate as outlined by Brown and Kellogg (13). Unfortunately, no satisfactory means for the carrying out of these two processes were at hand; consequently, some other method had to be devised. In developing such a method Kelley's (83) work on the hydrolysis of casein seemed to suggest a method whereby this assimilation factor could be eliminated. We know that casein is easily precipitated from neutral solutions by means of weak acids. This fact allows us to develop a method for the study of the ammonia production and ammonia

consumption process. In other words, should we employ this as a source of ammonifiable material we could eliminate the usual end point, ammonia, and instead use as a measure of activity what has remained behind unacted upon by the microorganisms. This can be carried out at certain specified intervals and in this manner the process may be studied periodically.

TABLE 15
The influence of sodium nitrate upon the ammonifying efficiency of soil bacteria

SOIL POSITION	NaNO ₃	BACILLUS SUBTILIS			BACILLUS TUMESCENTIS			BACILLUS MEGATERIUM			PER CENT RELATION TO CHECK	
		Ammonia Accumulation			Ammonia Accumulation			Ammonia Accumulation				
		M gm. N	M gm. N	Average M gm. N	M gm. N	M gm. N	Average M gm. N	M gm. N	M gm. N	Average M gm. N		
<i>per cent</i>												
1-2	None	30.50	31.30	30.90	100	11.32	9.02	10.17	100	10.22	9.92	10.07
3-4	0.010	36.26	36.56	36.41	117	10.22	10.64	10.43	102	8.62	9.12	8.87
5-6	0.025	36.11	36.01	36.06	116	10.04	9.92	9.98	98	10.24	9.04	9.64
7-8	0.050	35.48	35.06	35.27	113	12.42	14.40	13.41	132	8.84	8.64	8.74
9-10	0.100	30.40	29.78	30.09	99	13.52	9.62	11.57	113	7.04	8.70	7.87
11-12	0.250	28.20	26.80	27.50	89	7.30	8.04	7.67	75	8.44	9.92	9.18
13-14	0.500	25.90	26.06	25.98	84	5.44	6.34	5.89	57	7.22	8.54	7.88

SOIL POSITION	NaNO ₃	PSEUDOMONAS PUTIDA			BACILLUS MESENTERICUS			BACILLUS MYCOIDES			PER CENT RELATION TO CHECK	
		Ammonia Accumulation			Ammonia Accumulation			Ammonia Accumulation				
		M gm. N	M gm. N	Average M gm. N	M gm. N	M gm. N	Average M gm. N	M gm. N	M gm. N	Average M gm. N		
<i>per cent</i>												
1-2	None	54.50	58.00	56.25	100	14.50	13.70	14.10	100	13.30	13.30	100
3-4	0.010	59.58	57.68	58.63	103	18.03	17.58	17.80	111	14.92	14.52	14.72
5-6	0.025	52.58	51.62	52.10	92	15.00	14.92	14.96	105	14.20	13.92	14.06
7-8	0.050	59.38	59.36	59.37	106	15.58	14.68	10.13	70	14.37	14.80	14.58
9-10	0.100	64.54	62.00	63.27	116	15.40	12.20	13.80	92	12.40	12.17	12.28
11-12	0.250	54.70	49.00	51.85	92	12.92	12.22	12.57	80	10.25	10.25	79
13-14	0.500	35.66	37.06	36.36	64	12.90	12.95	12.93	81	8.94	8.84	8.89

It is upon these principles that the experimental work in this series has been based. In carrying out such an experiment, however, it is necessary to start with a good quality of casein. In order to secure this, it is necessary to reprecipitate that usually found on the market. The purification of the commercial casein was brought about as follows.

Commercial casein was dissolved in warm N/10 sodium hydrate and filtered through a double thickness of cotton. The filtrate was then acidified with weak acetic acid and the

casein precipitated. The supernatent liquid was then removed, the precipitate placed on a filter paper and washed free from acid with distilled water. The precipitate was then put in a flask and digested with ether for 24 hours. At the end of this time the ether was removed by means of 95 per cent alcohol which at the same time removed any sugars, etc. The casein was then dried over sulfuric acid in a water over at 100°C.

The resulting product was not of the same nitrogen analysis as the theoretical casein molecule, but was a marked improvement over the original material.

In carrying out the experimentation proper, enough of this purified material was dissolved in dilute N/10 sodium hydrate to give 100 mgm. of nitrogen per 100 cc. of solution; 100-cc. portions were then placed in 200-cc. Erlenmeyer flasks and varying quantities of sodium nitrate added as shown in table 16. Five grams of fresh Norfolk sandy loam were used as inoculating material, the flasks thoroughly shaken and then allowed to incubate for various intervals of time. The casein was precipitated from the 100 cc. of solution by means of

TABLE 16
The influence of NaNO₃ upon the ammonification and hydrolysis of casein

PERIOD OF INCUBA- TION	TREATMENTS							
	Series 1—No treat- ment		Series 2—10 Mgm. NaNO ₃		Series 3—50 Mgm. NaNO ₃		Series 4—100 Mgm. NaNO ₃	
	Accumulation of ammonia Mgm. N	Casein hydro- lyzed Mgm. N	Accumulation of Ammonia Mgm. N	Casein hydro- lyzed Mgm. N	Accumulation of Ammonia Mgm. N	Casein hydro- lyzed Mgm. N	Accumulation of Ammonia Mgm. N	Casein hydro- lyzed Mgm. N
days								
1	0.90	6.72	0.96	4.55	0.75	4.78	1.26	5.30
2	2.60	13.95	3.15	13.04	4.80	25.80	4.20	23.95
3	10.00	21.50	10.55	20.30	13.75	49.50	14.70	52.20
4	20.00	40.75	21.15	45.90	26.70	52.40	30.50	54.40
5	26.00	46.02	26.40	43.56	38.70	52.40	41.45	55.05
7	42.29	52.03	47.04	53.32	51.36	55.29	50.00	55.52

1 per cent acetic acid. This was filtered off and the total nitrogen in the same determined. Likewise the filtrate was analyzed for ammonia by the magnesium oxide method. The original solution of casein was precipitated and analyzed for total nitrogen and ammonia and the results calculated on the figures so obtained. It was likewise known that casein autohydrolyses. In order to take this into consideration an experiment was set up without inoculation. Bacterial infection was eliminated by means of toluol. The figures resulting from the autohydrolysis have also been taken into consideration in calculating the results.

In calculating the data the figures in the column headed "Accumulation of ammonia" represent the ammonia found in the filtrate after the casein has been precipitated by acetic acid. The column headed "Casein hydrolyzed" gives the amount of casein not precipitated by acetic acid after a given interval of time and represents that portion which has been acted upon by microorganisms. It will be seen by consulting table 16 that as the casein is acted upon by biological processes it goes into non-precipitable forms. That is to say,

as the interval of time increases between the first and subsequent analyses, we have less casein in solution. On the other hand, we have a corresponding increase in the hydrolyzed portion, with the formation of amino-acids and ammonia. The experiment was run in duplicate and the results given are the average of two determinations. The duplicates checked well.

It will be seen from a survey of table 16 that at the end of 24 hours very little action has taken place in the cultures. The untreated flasks have the greatest amount of casein hydrolyzed at this interval. At the end of 48 hours some marked changes have taken place in the various flasks. If we compare series 2 with the check flasks we find that no great change has taken place as a result of the presence of 10 mgm. of sodium nitrate. However, if we compare the third series with the untreated portion, we find that although the untreated portion is only slightly increased, the amount of casein hydrolyzed has increased very markedly. This is vastly out of proportion to the amount of ammonia accumulated if we conceive all the spilt ammonia as accumulating. It is very evident that something is happening which militates against the accumulation of ammonia. This is, likewise, true in the fourth and fifth series.

At the end of 72, 96, 120 and 144 hours this is again true. Where there is an increase in ammonia due to the action of sodium nitrate over that accumulating in the untreated series, the amounts of casein hydrolyzed at any given interval, as compared with the untreated portions, is much greater in all cases. Larger amounts are hydrolyzed earlier, with the higher concentration of the salt.

Of especial notice is the phenomenon in series 5. Although the ammonia accumulation is not as large as is present in any of the treated series the amount of casein hydrolyzed is much greater. At the end of 72 hours although only 12.3 mgm. of ammonia accumulated, the amount of casein hydrolyzed is greater than that hydrolyzed in the untreated portion in 168 hours. It is also greater than that hydrolyzed in series 4 in 120 hours.

It would seem then that the ammonia production-consumption theory was of great importance in ammonification studies.

We may accept the theory that sodium nitrate does actually increase biological activities to such an extent that an increased production of ammonia does not necessarily take place, although in fact the mineralization of the material has proceeded faster and to a greater extent. Such being the case, a new question arises, namely, as to the ultimate fate of this nitrogen and how soon it will again be available for plant growth. This question will be discussed later under another heading.

If, as has been pointed out, in some instances we receive an increase in the ammonia formed by applications of sodium nitrate, and in other cases no increase at all, may we not correlate this phenomenon with the increased or decreased recoveries of fertilizing material?

It has been observed that nitrate of soda when employed in extensive agri-

culture often gives returns much greater than can be accounted for by the quantity of plant-food actually supplied [Lipman (107)]. Moreover, it has been observed that the crop increase thus produced by nitrates may not be secured year after year, with ordinary applications, unless farm-yard manure or green manures be applied to offset the losses of organic matter and of nitrogen from the soil. For this reason sodium nitrate is believed by many farmers to be only a soil stimulant, hastening the depletion of soil fertility. Now, this belief is evidently due to an imperfect understanding of the observed facts, for it must be maintained that nitrate of soda is the most valuable of our nitrogenous fertilizers, and just as truly a plant-food as any incomplete fertilizer.

Where smaller returns of the nitrogen supplied as sodium nitrate are observed we may find an exhaustion, in the fact that there is some considerable conversion of applied nitrogen into protein forms for the microbial cell, resulting in a lesser quantity of nitrate being available for plant growth.

It does not follow, however, that because the influence of sodium nitrate upon soil microorganisms is being studied entirely from the nitrogen standpoint, its application does not bear an important relation to the mineral elements of plant-food, i.e., K, P, Mg, Fe, S, and silicon. Potter and Snyder (143), Ramann (140), Van Suchtelen (199), have all reported data to show the influence of fertilizers on carbon-dioxide production in soils. In the degradation of the organic matter carbon-dioxide is liberated. An increase in ammonia production necessarily results in a greater amount of carbon dioxide. Remembering that carbon dioxide is an excellent solvent and must play an important rôle in the weathering of rock fragments, we receive a hint as to the possible influence of sodium nitrate in indirectly rendering available the mineral plant-food elements of the soil. The breaking down of the soil humus by bacteria is accompanied by the formation of various organic acids which also react on the constituents of the rock fragments.

Series 10. Is the stimulating action of sodium nitrate due to the sodium or the nitrate portion of the molecule?

We must not leave the action of sodium nitrate upon the ammonifying group without first ascertaining whether its action is due to its sodium or its nitrate ions.

Greaves (54) in his researches says that he has good reason to believe that the anion is the most influential part of the salt, the cation being only of minor significance. His work is based upon the ammonification of dried blood. In order to see if the same thing held true with regard to sodium nitrate upon the ammonification of cottonseed meal, another series of experiments was set up, employing the four sodium salts, NaCl , Na_2CO_3 , NaNO_3 , and Na_2SO_4 , with a constant amount of sodium, and the consequential variation of the anion. The beaker method was used and the soil was the Norfolk sandy loam. The results are given in table 17.

It is quite evident from an analysis of the table that the acid radical is of much importance in regulating the ammonification of cottonseed meal. With increases in the amounts of all four salts are obtained corresponding increases in the amount of ammonia formed. The CO_3^{2-} ion seems to be the most toxic of the four anions, with the chloride next, and the nitrate and sulfate in the order named. Parenthetically, it may be pointed out that in only one case, that of Na_2CO_3 , was there any marked toxic influence present even at a concentration of 50,000 pounds per acre.

TABLE 17
The influence of the anion upon the ammonification of cottonseed meal

SOIL PORTION	POUNDS PER ACRE OF SALTS	NaNO ₃ SERIES			PER CENT RELATION TO CHECK	NaCl SERIES			PER CENT RELATION TO CHECK		
		Ammonia accumulation				Ammonia accumulation					
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N			
1-2	None	29.32	30.94	30.13	100	22.80	22.80	22.80	100		
3-4	300	42.70	42.60	42.65	140	26.10	22.60	24.35	106		
5-6	900	54.17	53.94	54.05	179	20.00	26.00	23.00	105		
7-8	1,500	62.20	55.40	58.80	195	31.00		31.00	136		
9-10	2,500	60.20	59.10	59.65	198	33.00	33.00	33.00	144		
11-12	5,000	56.60	57.08	56.84	188	33.80	33.80	33.80	145		
14-14	10,000	63.13	61.98	62.54	207	38.60	42.40	40.50	177		
15-16	30,000	69.30	72.40	70.85	235	70.60	69.00	69.80	306		
17-18	50,000	65.20	68.30	67.20	229	51.60	54.00	52.80	231		

SOIL PORTION	POUNDS PER ACRE OF SALTS	Na ₂ CO ₃ SERIES			PER CENT RELATION TO CHECK	Na ₂ SO ₄ SERIES			PER CENT RELATION TO CHECK		
		Ammonia accumulation				Ammonia accumulation					
		Mgm. N	Mgm. N	Average mgm. N		Mgm. N	Mgm. N	Average mgm. N			
1-2	None	22.80	22.80	22.80	100	12.00	12.00	12.00	100		
3-4	300	25.00	28.00	26.50	115	15.40		15.40	128		
5-6	900	58.00	43.00	50.50	221	12.00	12.00	12.00	100		
7-8	1,500	52.40	52.40	52.40	227	8.40	6.80	7.60	73		
9-10	2,500	64.10	61.60	62.85	279	8.80	8.90	8.85	83		
11-12	5,000	78.40	64.00	71.20	312	7.40	8.80	8.20	82		
13-14	10,000	65.60	66.00	65.80	289	14.00	10.80	14.40	180		
15-16	30,000	39.60	41.20	40.40	177	20.00	22.80	21.90	182		
17-18	50,000	12.00	12.60	12.30	43	26.60	31.60	29.10	242		

PART III

The influence of sodium nitrate upon the nitrification process

Of all the nitrogen compounds yet investigated nitrates are the best, and in natural conditions probably the only nitrogenous food for the non-leguminous plants. This probability takes form from the fact that most soils are supplied with two species of soil bacteria which rapidly transform the ammonia formed by the ammonifying organisms into nitrates, the process, in the broad sense, being termed nitrification. More specifically, however, it applies to the transformation of nitrite to nitrate by *Nitrobacter*. In this text the term nitrification is used to describe the entire process of mineralization from ammonia to nitrates.

As this process is of the utmost importance for the nutrition of plants it is, therefore, of special significance to know just what the influence of additional nitrates would have upon the process.

Investigators have given us some insight into the action of sodium nitrate upon the nitrification process. For instance, Lipman and Brown (109) state that sodium nitrate markedly enhances nitrification. This also seems to be the impression one would get from the work of Sackett (159) and Wöhltmann (226). Miyake (127), using the data of Lipman and Brown (109) and Warington (214) proves mathematically that nitrification is an auto-catalytic reaction, i.e., the presence of the end point seems to hasten the process along. On the other hand, the work of Brown and Minges (14) Deherain (25), Kelley (81), and Fred and Graul (38) seems to indicate that nitrification is not benefited, but is depressed by the presence of sodium nitrate. Warington says that nitrates do not affect the nitrification of soil humus, but that $(\text{NH}_4)_2\text{SO}_4$ does, because it becomes nitrified rather than the soil's organic matter.

It will be seen, therefore, that the opinions with regard to its action are conflicting, and it is very desirable to have further evidence on the subject.

In doing so the writer has studied the influence of sodium nitrate in five different types of soil, with three sources of nitrifiable material, the data being recorded in the following pages.

The method used in studying the process was the beaker method before described (108). Organic matter was supplied in amounts equivalent to 21 mgm. of nitrogen. Calcium carbonate was added to neutralize soil acidity and leave an excess equal to 0.02 per cent. The incubation period was 28 days at a temperature of 20°C. Moisture conditions were 50 per cent of the water-holding capacity of the soil.

The method of extracting the nitrates was that worked out by Allison (2) and the method of determining the same was that proposed by Allen (1). This exception, however, is to be noted—the quartz distilling tubes recommended by him were not used. Instead, a Kjeldahl rack with block tin tubes supplied with extra large-sized traps were employed. An average error of 0.5 cc. of N/50 acid was experienced with this modification. As

the work is comparative rather than absolute this error is of little importance, being involved in every determination.

Series 11. The influence of sodium nitrate upon the nitrification of ammonium sulfate

The first experiments were carried out to test the effect of sodium nitrate upon the nitrification of ammonium sulfate. Five soils were used. The fertilizer additions, nitrates accumulating and calculated data are given in table 18. It will be seen upon consulting the table that sodium nitrate seems to depress the accumulation of nitrates in every one of the soils tested. The greatest reaction seems to have taken place in the Norfolk sandy loam. In this soil applications of sodium nitrate of only 50 pounds per acre decreased the accumulation of nitrates 7 per cent. With succeeding increments a gradual depressed action is manifested being 30 per cent below normal at a concentration of 300 pounds per acre and reaching 82 per cent below normal at a concentration of 1500 pounds per acre. It seems as if the nitrification of this material is stopped entirely when 2500 pounds per acre are supplied as this type of soil.

A similar phenomenon is to be seen also in the Penn clay loam. In this soil the depression is not so large as in the previous soil. Fifty pounds per acre decreases the accumulation only 3 per cent, 500 pounds per acre 10 per cent and 1500 pounds 46 per cent below the normal nitrifying power of the soil. A small amount of nitrification, 5 per cent, was experienced at a concentration equal to 2500 pounds per acre.

In the Wooster silt loam decreased accumulation of nitrates is not to be seen until a concentration equivalent to 500 pounds per acre is reached. From here on, however, there is a gradual decrease in the nitrate accumulation. The toxic action of sodium nitrate is not nearly as great in this soil as was the case in the Penn clay loam. Even at a concentration of 2500 pounds per acre the nitrate accumulation is only 53 per cent below normal. At double this concentration no nitrification took place, however.

Comparing the Carrington loam with these three soils we find that sodium nitrate has apparently not affected the nitrification of ammonium sulfate to any great extent. There seems to be no decrease in the amount of nitrates accumulating below normal until a concentration of 2500 pounds per acre is reached. The depression at this concentration is only 7 per cent below normal. Doubling this concentration, however, decreases the nitrifying power to about 40 per cent of normal.

The accumulation of nitrates under normal conditions in the Sierra sandy loam and the Norfolk sandy loam is practically identical. However, the action of sodium nitrate upon the nitrification of ammonium sulfate is by no means the same. In the Sierra sandy loam there is a strong indication of a stimulative action. At any rate, there is no decreased accumulation of

The influence of NaNO_3 upon the nitrification of ammonium sulfate

SOIL PORTION	SPECIAL TREATMENT	CARRINGTON LOAM				NORFOLK SANDY LOAM				PENN CLAY LOAM			
		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation	
		Mg.m. N	Average Mg.m. N										
1-2	Nothing.....	8.60	8.61	8.60	8.61	4.01	4.01	4.01	4.01	7.84	7.84	7.84	7.84
3-4	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	23.75	23.85	23.80	23.80	15.20	15.20	10.00	11.85	11.49	11.49	11.45	11.45
5-6	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	24.58	24.68	24.63	24.63	16.03	15.62	10.2	11.66	11.49	11.57	11.56	11.56
7-8	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	24.58	24.60	24.59	24.59	15.99	15.99	11.00	11.60	11.49	11.54	11.53	11.53
9-10	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	26.58	26.99	26.78	26.78	18.16	18.16	1.19	10.91	10.85	10.88	6.87	6.87
11-12	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	27.11	27.35	27.23	27.23	18.63	18.63	1.48	10.10	10.36	10.20	10.26	10.26
13-14	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	28.34	28.50	28.42	28.42	19.80	19.80	1.54	10.1	12.52	12.52	12.52	12.52
15-16	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	29.71	29.93	29.82	29.82	21.66	21.66	1.13	10.9	12.90	12.90	12.90	12.90
17-18	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	30.98	31.30	30.90	30.90	23.76	23.76	15.16	10.0	12.80	12.80	8.79	8.79
19-20	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	32.71	32.33	32.52	32.52	21.92	21.92	1.41	10.1	12.37	12.37	12.12	12.12
21-22	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	38.90	37.53	38.21	38.21	21.91	21.91	1.47	9.3	15.30	15.31	15.30	15.30
23-24	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	45.51	46.04	45.77	45.77	37.17	37.17	8.03	6.2	23.26	23.26	23.26	23.26
25-26	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	64.60	63.86	64.23	64.23	35.63	35.63	46.00	0	46.22	45.92	46.12	42.11
										-3.17	0	63.74	63.54
											0	63.55	63.79
												64	64

SOIL PORTION	SPECIAL TREATMENT	SIERRA SANDY LOAM				WOOSTER SILT LOAM			
		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation		Nitrate accumulation	
		Mg.m. N	Average Mg.m. N	Mg.m. N	Average Mg.m. N	Mg.m. N	Average Mg.m. N	Mg.m. N	Average Mg.m. N
1-2	Nothing.....	5.15	5.15	5.15	5.15	7.43	7.43	7.43	7.43
3-4	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	12.43	12.74	12.58	12.58	8.05	8.05	7.80	7.80
5-6	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	13.24	13.57	13.40	13.40	12.87	12.87	7.72	7.72
7-8	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	14.30	15.00	14.95	14.95	9.80	9.80	8.36	8.36
9-10	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	15.38	15.11	15.24	15.24	10.09	10.09	7.86	7.86
11-12	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	15.46	15.15	15.24	15.24	10.09	10.09	6.74	6.74
13-14	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	16.65	17.30	16.97	16.97	11.82	11.82	7.36	7.36
15-16	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	17.22	17.52	17.37	17.37	12.12	12.12	5.22	5.22
17-18	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	18.11	17.64	17.86	17.86	12.73	12.73	1.79	1.79
19-20	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	26.74	26.18	26.06	26.06	24.91	24.91	2.92	2.92
21-22	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	26.97	26.49	26.66	26.66	24.69	24.69	2.79	2.79
23-24	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	26.97	26.49	26.66	26.66	24.69	24.69	2.79	2.79
25-26	100 mgm. $(\text{NH}_4)_2\text{SO}_4 + \text{NaNO}_3$ -	26.97	26.49	26.66	26.66	24.69	24.69	2.79	2.79

nitrates to speak of until a concentration equivalent to 1500 pounds per acre of the salt is present. At this point the depression is about 30 per cent below normal. Beyond this point there is a rapid and pronounced decreased activity. In attempting to explain the difference in the action of the two sandy soils we must remember that the Sierra sandy loam is from a semi-arid region having a greater amount of sodium salts in its soil solution.

C. B. Lipman (104) has published his so-called antagonism theory in which he finds that the addition to soils toxic to one form of the three sodium salts, NaCl , Na_2CO_3 and Na_2SO_4 , of quantities of any other of these salts will result in an improved condition of the soil flora. If these three salts can aid one another, why cannot sodium nitrate do the same thing? Let us suppose the Sierra sandy loam has one of the salts as a toxic factor. Why would not the applications of NaNO_3 aid in the rearrangement of the soil solution in a manner that would help the nitrifying group? In the Norfolk sandy loam, however, no abundance of so-called alkali salts could accumulate and this explanation is not tenable.

It seems also from a consideration of the data that there is a marked qualitative difference in the resistance of the different strains of the nitrifying organisms to applications of sodium nitrate. For instance, in the Wooster silt loam, having 20 per cent of moisture in the system, the depressed action at a concentration of 1500 pounds per acre is 36 per cent below normal, whereas in the Penn clay loam with 25 per cent of moisture in the system the decreased accumulation is 46 per cent below normal. In the Carrington loam, on the other hand, with the same amount of moisture no depressive action at all is to be noted. Of course, the concentration of the soil solution itself would have something to do with such a generalization, but nevertheless, differences of 50 per cent could hardly be explained away in such a manner. Correlating the phenomenon in the above experiments with actual field conditions, it would seem that the beneficial use experienced with a mixture of sodium nitrate and ammonium sulfate might be explained on the grounds that sodium nitrate decreased the mineralization of the latter until the time was opportune for its assimilation by the plant, or again, the selective action of crops for ammonia, as has been noted previously (164), might enter in, the NaNO_3 , depressing the nitrate formation, might permit the ammonia, as such, to become assimilated.

Series 12. The influence of sodium nitrate upon the nitrification of dried blood nitrogen

This series consisted of a study of the effect of sodium nitrate upon the nitrification of dried blood. The data are to be found in table 19. The action of sodium nitrate upon the nitrification of dried blood was not studied in the Norfolk sandy loam in this series.

Considering first the nitrification of the dried blood itself in this series, we

TABLE 19
The influence of sodium nitrate upon the nitrification of dried blood

SODIUM NITRATE PER 100 LBS.	SPECIAL TREATMENT	CARRINGTON LOAM				SANDY SANDY LOAM			
		Nitrate accumulation in mgm. N		Nitrate accumulation in mgm. N		Nitrate accumulation in mgm. N		Nitrate accumulation in mgm. N	
		Mgns. no treat- ment added NaNO ₃	Average N	Mgns. no treat- ment added NaNO ₃	Average N	Mgns. no treat- ment added NaNO ₃	Average N	Mgns. no treat- ment added NaNO ₃	Average N
1-2	Nothing.....	5.95	6.26	6.10	6.13	6.13	6.13	6.13	6.13
3-4	1731 mgm. D. B. + NaNO ₃ = 50 lbs. per acre.....	21.33	21.49	21.41	15.31	100	20.35	19.95	20.15
5-6	1731 mgm. D. B. + NaNO ₃ = 100 lbs. per acre.....	21.49	22.31	21.90	15.80	15.37	100	19.95	20.13
7-8	1731 mgm. D. B. + NaNO ₃ = 200 lbs. per acre.....	22.98	22.63	22.80	16.70	15.87	103	20.04	13.91
9-10	1731 mgm. D. B. + NaNO ₃ = 300 lbs. per acre.....	23.14	23.68	23.41	17.31	15.29	100	19.10	18.48
11-12	1731 mgm. D. B. + NaNO ₃ = 500 lbs. per acre.....	24.24	24.34	24.29	18.19	15.16	99	20.67	20.81
13-14	1731 mgm. D. B. + NaNO ₃ = 700 lbs. per acre.....	25.34	25.17	25.25	19.15	14.87	97	20.95	21.58
15-16	1731 mgm. D. B. + NaNO ₃ = 900 lbs. per acre.....	28.98	25.80	25.89	19.79	14.25	93	20.01	20.42
17-18	1731 mgm. D. B. + NaNO ₃ = 1,200 lbs. per acre.....	28.50	28.50	28.50	22.40	14.55	94		
19-20	1731 mgm. D. B. + NaNO ₃ = 1,500 lbs. per acre.....	29.71	29.71	29.71	23.61	14.06	91	20.65	21.08
21-22	1731 mgm. D. B. + NaNO ₃ = 2,500 lbs. per acre.....	33.10	33.76	33.46	27.36	12.21	80	27.95	28.15
23-24	1731 mgm. D. B. + NaNO ₃ = 5,000 lbs. per acre.....	40.15	40.15	40.15	34.05	4.15	26	35.16	34.58
25-26	1731 mgm. D. B. + NaNO ₃ = 10,000 lbs. per acre.....	63.31	63.31	63.31	57.21	1.16	0	43.32	42.75

SOIL TREATMENT	PENN CLAY LOAM			WOOSTER SILT LOAM		
	NITRATE ACCUMULATION			NITRATE ACCUMULATION		
	Mg/m. N AVERAGE	Mg/m. N MILIMS NO TREAT.	Mg/m. N MILIMS NO TREAT.	Mg/m. N AVERAGE	Mg/m. N MILIMS NO TREAT.	Mg/m. N MILIMS NO TREAT.
1-2 Nothing.....	8.96	8.96	8.96	6.82	6.82	6.82
3-4 1731 mgm. D. B. + NaNO ₃ = 50 lbs. per acre.....	23.47	23.71	23.59	14.63	100	10.50
5-6 1731 mgm. D. B. + NaNO ₃ = 100 lbs. per acre.....	23.47	23.47	23.47	14.51	14.09	97
7-8 1731 mgm. D. B. + NaNO ₃ = 300 lbs. per acre.....	23.34	23.10	23.22	14.26	13.52	92
9-10 1731 mgm. D. B. + NaNO ₃ = 500 lbs. per acre.....	24.91	25.15	25.03	16.07	14.09	97
11-12 1731 mgm. D. B. + NaNO ₃ = 700 lbs. per acre.....	26.12	27.12	26.67	17.71	14.57	99
13-14 1731 mgm. D. B. + NaNO ₃ = 900 lbs. per acre.....	27.16	28.00	27.58	18.62	14.38	98
15-16 1731 mgm. D. B. + NaNO ₃ = 1,200 lbs. per acre.....	29.95	26.78	26.86	17.90	13.80	87
17-18 1731 mgm. D. B. + NaNO ₃ = 1,500 lbs. per acre.....	28.16	30.50	29.37	20.41	12.41	84
19-20 1731 mgm. D. B. + NaNO ₃ = 2,500 lbs. per acre.....	29.00	31.52	30.26	21.20	12.05	82
21-22 1731 mgm. D. B. + NaNO ₃ = 5,000 lbs. per acre.....	32.65	31.85	32.24	23.28	6.40	42
23-24 1731 mgm. D. B. + NaNO ₃ = 10,000 lbs. per acre.....	37.09	37.75	37.42	28.46	-0.34	0
25-26 1731 mgm. D. B. + NaNO ₃ = 10,000 lbs. per acre.....	62.26	61.91	62.18	53.22	-4.94	0

find the Carrington loam nitrifying this material as effectively as ammonium sulfate. This was, likewise, true to a large measure with the nitrification of this material in the Penn loam soil. The Sierra sandy loam, on the other hand, nitrified dried blood to twice the extent that it did ammonium sulfate. The Wooster silt loam had very little ability to nitrify dried blood.

In the action of sodium nitrate upon the nitrification of this material we again find in every case a decreased accumulation of nitrates. In the Carrington loam this does not take place until a concentration of 500 pounds per acre is reached. From this point on, however, we note a gradual decrease in the accumulation of nitrates.

In this series the effect of sodium nitrate upon the accumulation of nitrates is not so seriously affected as was the case in the previous series. That is to say, the toxic action does not present itself in such magnitude as was the case where ammonium sulfate was the nitrifiable material.

The Wooster silt loam is the only soil in this series to exhibit an enhanced activity as a result of the presence of soluble nitrates. Because of the small amount of nitrification and the difference between parallel determinations it is even questionable whether this is actually the case, or only apparent from a mathematical standpoint.

The nitrification of dried blood in the Sierra loam is discouraged by the application of sodium nitrates in this series, in distinction to the previous effect. The Penn clay loam, likewise, showed a decrease in accumulation of nitrates as a result of the presence of sodium nitrate in this series, as was the case in the previous one.

The accumulation of nitrates as compared with the untreated soils is much greater in this series than under similar conditions in the previous series. To state the matter in other words, quantities of soluble nitrates do not seem to discourage the accumulation of nitrates from the nitrification of dried blood nitrogen as they did from ammonium sulfate. Perhaps this may be explained on the grounds of the concentration of the soil solution. When ammonium sulfate is added to the soil we have, of course, a considerable increase in the salt content of the soil solution. The concentration, i.e., osmotic concentration, is further enhanced by applications of sodium nitrate. Moreover, as ammonium sulfate rapidly changes into nitrates this ion likewise increases the osmotic pressure. The summation of these three probably was great enough seriously to discourage the nitrate building bacteria. With dried blood, on the other hand, not only is the initial concentration small but the increase in the total concentration arrives more gradually because of the slower nitrifiability of the substance. Thus the organisms have a chance to adapt themselves to their new environment.

Knowing that cottonseed meal ammonifies even more slowly than dried blood, we would expect a lesser depression in the accumulation of nitrates from high applications of sodium nitrate than from dried blood. To test this out another series was set up, the results of which are given below.

Series 13. The influence of sodium nitrate upon the nitrification of cottonseed meal nitrogen

From an examination of table 20 it will be seen that all soils nitrify cottonseed meal to a lesser extent than dried blood or ammonium sulfate. The soil nitrifying this material to the greatest extent was the Norfolk sandy loam.

Considering the nitrification process in the order in which it was affected in the various soils we find that marked stimulation occurred in three of the five soils, viz., the Penn clay loam, the Sierra sandy loam, and the Wooster silt loam. The Sierra sandy loam shows a marked stimulating phenomenon in all but two cases, the maximum amount accumulating with a concentration of 700 pounds per acre. Curiously enough, there was no nitrification of cottonseed meal in this soil at a concentration of 5000 pounds, although ammonium sulfate was nitrified to an extent of about 50 per cent below normal under the same conditions.

The Carrington loam's ability to nitrify cottonseed meal was not affected to any extent by applications of sodium nitrate until a concentration of 2500 pounds per acre was at hand. As was the case in all of the soils no nitrification took place at a concentration equivalent to 10,000 pounds per acre. It would seem also that the theory advanced in the former series was more than operating here, to allow the accumulation of relatively larger proportions of nitrates. If we consider the effect of nitrate of soda upon the nitrification of the three nitrogen-bearing materials we note that the greatest decrease in the nitrate accumulations was experienced first with ammonium sulfate as the nitrifiable material, next with dried blood and lastly with cottonseed meal. The greatest decrease is found in sandy soils, silt soils and loam soils, in the order named.

From Kelley's (83) work we are led to believe that nitrification is at its maximum at the end of two weeks' time. If such is the case we would probably expect a more decided depression in the accumulation of nitrates due to applications of sodium nitrate. In order to test this out and at the same time try to explain why sodium nitrate did not decrease the accumulation of nitrates in the Carrington silt loam, a new series of experiments were instituted designed to determine the nitrates formed from the nitrification of ammonium sulfate, dried blood and cottonseed meal at the end of 8, 16, 24, and 32 days.

Series 14. The influence of sodium nitrate upon the accumulation of nitrates derived from the nitrification of ammonium sulfate at various intervals of time

In testing out the influence of time upon the accumulation of nitrates four applications of nitrate of soda were used, viz., 50, 100, 300, and 500 pounds per acre. Otherwise the method of study was identical with the previous work.

Table 21 records the data on the accumulation of nitrates from the nitrification of ammonium sulfate at different periods of time. At the end of the

TABLE 20
The influence of sodium nitrate upon the nitrification of cottonseed meal

LAB. NO.	SPECIAL TREATMENT	CARRINGTON LOAM			NORFOLK SANDY LOAM			PENN CLAY LOAM		
		Nitrate accumulation		Mg.m. N	Nitrate accumulation		Mg.m. N	Nitrate accumulation		Mg.m. N
		Mg.m. N	Average Mg.m. N		Mg.m. N	Average Mg.m. N		Mg.m. N	Average Mg.m. N	
1-2	Nothing	5.95	6.26	6.10	9.91	9.91	100	14.60	14.31	14.45
3-4	300 mgm. C. S. M. + NaNO ₃	15.96	16.06	16.01	9.91	2.79	2.79	11.66	11.66	100
5-6	300 mgm. C. S. M. + NaNO ₃	16.31	16.64	16.47	10.37	9.94	100	13.77	14.59	14.11
7-8	300 mgm. C. S. M. + NaNO ₃	16.49	17.03	16.76	10.66	9.83	100	14.41	14.43	14.42
9-10	300 mgm. C. S. M. + NaNO ₃	18.25	18.18	18.19	12.09	10.07	101	15.80	15.59	15.69
11-12	300 mgm. C. S. M. + NaNO ₃	18.85	19.13	18.99	12.89	9.86	100	16.25	15.70	15.97
13-14	300 mgm. C. S. M. + NaNO ₃	19.49	19.77	19.79	13.53	9.27	94	17.67	16.08	16.87
15-16	300 mgm. C. S. M. + NaNO ₃	21.61	21.77	21.74	15.64	10.10	101	18.02	18.07	18.05
17-18	300 mgm. C. S. M. + NaNO ₃	24.03	24.24	24.12	18.02	10.27	102	19.18	19.16	19.10
19-20	300 mgm. C. S. M. + NaNO ₃	25.23	24.83	25.03	18.93	9.83	100	20.25	20.14	20.69
21-22	300 mgm. C. S. M. + NaNO ₃	30.62	30.30	30.85	24.40	9.35	95	23.16	23.20	23.76
23-24	300 mgm. C. S. M. + NaNO ₃	39.81	39.90	39.85	33.75	4.85	50	28.90	29.00	29.25
25-26	300 mgm. C. S. M. + NaNO ₃	63.31	63.31	63.31	57.70	-1.16	0	43.59	44.46	44.02

LAB. NO.	SPECIAL TREATMENT	SEWERA SANDY LOAM			WOOSTER SILT LOAM			
		Nitrate accumulation		Mg.m. N	Nitrate accumulation		Mg.m. N	
		Mg.m. N	Average Mg.m. N		Mg.m. N	Average Mg.m. N		
1-2	Nothing	6.22	6.08	6.15	8.89	8.89	100	9.32
3-4	300 mgm. C. S. M. + NaNO ₃	13.14	14.95	15.04	14.10	10.30	115	9.44
5-6	300 mgm. C. S. M. + NaNO ₃	15.71	16.11	15.91	9.76	9.53	107	10.04
7-8	300 mgm. C. S. M. + NaNO ₃	16.22	16.27	16.29	10.14	9.67	108	11.86
9-10	300 mgm. C. S. M. + NaNO ₃	16.74	16.67	16.70	10.55	9.62	108	11.62
11-12	300 mgm. C. S. M. + NaNO ₃	18.37	18.36	18.36	12.21	9.68	108	12.42
13-14	300 mgm. C. S. M. + NaNO ₃	20.02	20.48	20.25	14.10	10.30	115	10.84
15-16	300 mgm. C. S. M. + NaNO ₃	20.50	19.76	20.18	14.03	9.46	106	11.90
17-18	300 mgm. C. S. M. + NaNO ₃	21.59	21.05	21.32	13.17	9.45	106	14.92
19-20	300 mgm. C. S. M. + NaNO ₃	22.54	22.54	22.54	16.39	9.29	104	15.78
21-22	300 mgm. C. S. M. + NaNO ₃	26.64	26.51	26.57	20.42	8.87	100	18.70
23-24	300 mgm. C. S. M. + NaNO ₃	36.30	36.23	36.21	20.06	7.14	81	32.06
25-26	300 mgm. C. S. M. + NaNO ₃	44.03	44.03	43.51	43.77	3.06	0	32.80

TABLE 21
The influence of sodium nitrate upon the nitrification of ammonium sulfate at various intervals

eighth day the cultures having ammonium sulfate, and this material plus sodium nitrate equivalent to 50 and 100 pounds per acre, all had less nitrates in the system than a similar portion of untreated soil. Those portions, however, having nitrate of soda equal to 300 and 500 pounds per acre registered a decided increase over the untreated cultures.

On the sixteenth day, however, this stimulative influence seems to have been lost, as a decided decreased accumulation of nitrates was found in all cases, varying from 6 per cent to 48 per cent below the normal nitrification.

When the soils were analyzed for nitrates at the end of the twenty-fourth day this relative depressed accumulation was considerably narrowed and at 50 pounds per acre an actual stimulation of 2 per cent is to be noted. The maximum decrease here is only 18 per cent below the normal. It would seem that from the sixteenth to the twenty-fourth day a marked stimulation to the nitrifying group has occurred, the total nitrates formed being enough to approach the accumulation in the check cultures and to register an enhanced effect.

At the end of the experiment we find a slight increase over the untreated portions in all cases. No decrease in the nitrate accumulation is to be noted in any case which is in confirmation of the previous results.

Series 15. The influence of sodium nitrate upon the accumulation of nitrates derived from the nitrification of cottonseed meal

Concerning the effect of sodium nitrate upon the nitrification of cottonseed meal we must come to the same conclusion as was reached in series 20. At the end of 8 days no accumulation of nitrates at all is to be seen.

In sixteen days the applications of sodium nitrate equivalent to 300 and 500 pounds per acre have decreased the accumulation of nitrates 89 and 75 per cent, respectively. On being analyzed for nitrates at the end of the twenty-fourth day the data show that the depressed accumulation is materially lessened, as was the case with ammonium sulfate. At the finish of the experiment the resulting action of sodium nitrate upon nitrification of cottonseed meal is nil.

Series 16. The influence of sodium nitrate upon the accumulation of nitrates derived from the nitrification of dried blood

The data concerning the influence of sodium nitrate upon the nitrification of dried blood are given in table 21A. The greatest nitrification of all the materials was at the end of the experiment. The increase in nitrate accumulation from the twenty-fourth to the thirty-second day was small indeed.

An objection to the figures tabulated above can be raised on the grounds that nitrates as well as nitrites are reduced by this reduction method. However, this factor is shown to be insignificant under another experiment.

TABLE 21A
The influence of sodium nitrate upon the nitrification of dried blood at various intervals

In attempting to explain the decreased nitrate accumulation due to the presence of sodium nitrate we must again consider the consumption theory as previously explained. Nitrates, as well as ammonia, are readily converted into insoluble forms. Due to the addition of sodium nitrate as well as to nitrification, a marked increase takes place in the numbers of the microflora of the soil. These organisms necessarily need nitrogen for their life process and as nitrates, and even the ammonia from the nitrifying substances are present in excess, they use it for their own life processes. Fred and Graul (38) have given us a neat piece of experimentation on this point, as one will see by consulting the data on page 333 of their article.

We can also tell to a considerable degree just how far this consumption phenomenon is being carried out and when the toxicity factor enters into these experiments, if we titrate the ammonia, not converted into nitrate, which is driven off from the extracted nitrate solution in the carrying out of this reduction method. Although the figures thus obtained cannot be quantitative because of absorption of this ammonia by the soil, it is nevertheless extremely indicative of the point when excess nitrates seem to discourage the nitrifying organisms from converting ammonia into nitrate.

From an examination of the data recorded in table 22 there are to be seen marked differences at the points which sodium nitrate inhibits the nitrifiability of ammonium sulfate, dried blood or cottonseed meal. Toxicity commences to manifest itself in the Carrington loam at a concentration of about 1500 pounds per acre, where dried blood and cottonseed meal are the nitrifiable materials. In the Norfolk sandy loam the depressed action of sodium nitrate upon the nitrifiers is present at smaller concentrations, viz., 500 pounds per acre. When cottonseed meal, however, is the nitrifiable material no toxicity occurs until a concentration of 5000 pounds per acre is present. The toxicity of sodium nitrate to the nitrification of ammonium sulfate was not studied in this connection. Its toxicity to the mineralization of dried blood is evident at a concentration of 2500 pounds per acre and to that of cottonseed meal at 2500 pounds per acre. In the Sierra sandy loam no toxic action is noticed to affect the nitrification of cottonseed meal until a concentration of 5000 pounds per acre is reached. It is here just one-half as great as it is at double this concentration. Noticeable toxicity to the mineralization of dried blood starts quite early, viz., at a concentration of 1200 pounds per acre. Increasing the amount increases the toxic effect. No discouraging action to the nitrifying organisms is present in the Wooster silt loam until a concentration of over 500 pounds per acre of sodium nitrate is reached, with ammonium sulfate and dried blood as the source of nitrogenous material. The maximum toxicity in the Wooster silt loam in so far as the nitrification of dried blood and cottonseed meal is concerned, occurs at a concentration of 5000 to 10,000 pounds per acre.

Of course, in using the reduction method nitrites are reduced, if present, as well as nitrates and the ammonium formed is necessarily the sum of the

TABLE 21a
The influence of sodium nitrate upon the nitrification of cottonseed meal at various intervals

LAB. NO.	SPECIAL TREATMENT	PER CENT RELATION TO MINES NANO ₃ ADD'D		PER CENT RELATION TO MINES NANO ₃ ADD'D		PER CENT RELATION TO MINES NANO ₃ ADD'D		PER CENT RELATION TO MINES NANO ₃ ADD'D		PER CENT RELATION TO MINES NANO ₃ ADD'D		
		8 DAYS	16 DAYS	24 DAYS	32 DAYS	Mg.m. N	Average Mg.m. N	Mg.m. N	Average Mg.m. N	Mg.m. N	Average Mg.m. N	
-2	Nothing.....	6.67	6.77	7.92	7.73	8.38	8.34	8.59	8.55	8.57	8.57	
-4	300 mgm. C. S. M. + NaNO ₃ = 50	6.84	7.05	6.94	0.27	100	14.15	14.31	14.23	6.50	100	17.55
-6	300 mgm. C. S. M. + NaNO ₃ = 100	7.03	6.76	6.89	0.11	-0.30	0	14.50	14.16	14.33	6.00	5.18
-8	300 mgm. C. S. M. + NaNO ₃ = 100 pounds per acre.....	7.50	6.87	7.18	0.41	-0.43	0	15.25	15.28	15.24	7.55	6.72
-10	300 mgm. C. S. M. + NaNO ₃ = 300 pounds per acre.....	8.44	8.44	8.44	1.67	-0.32	0	15.49	15.67	15.58	7.85	5.86
-12	300 mgm. C. M. S. + NaNO ₃ = 500 pounds per acre.....	9.71	9.71	9.71	2.94	-0.21	0	15.95	15.40	15.67	9.44	7.79

two. Kelley (83) considers that a considerable amount of nitrites is formed when amounts of organic matter are present in excess of what he proposes as a standard, i.e., 10 mgm. of nitrogen per 100 gm. of soil. As approximately twice that figure was used in these experiments it is evident, if his conclusions are tenable, that large amounts of nitrites have been formed and are recorded as nitrates.

At the same time while we consider this question of nitrite formation it is not out of place to note, if possible, whether sodium nitrate will discourage the action of *Nitrobacter* and allow the production of considerable quantities of nitrites. Kelley has noted that certain alkali salts seem to cause such a phenomenon. It is of importance to know if considerable amounts of nitrites are formed because of their extreme toxicity to plants.

TABLE 22
Nitrogen as ammonia not nitrified in the presence of sodium nitrate

NaNO ₃ per acre	CARRINGTON LOAM			NORFOLK SANDY LOAM			PENN CLAY LOAM			SIERRA SANDY LOAM			WOOSTER SILT LOAM		
	From (NH ₄) ₂ SO ₄	From dried blood	From cotton seed	From (NH ₄) ₂ SO ₄	From dried blood	From cotton seed	From (NH ₄) ₂ SO ₄	From dried blood	From cotton seed	From (NH ₄) ₂ SO ₄	From dried blood	From cotton seed	From (NH ₄) ₂ SO ₄	From dried blood	From cotton seed
lbs.	mgm.	mgm.	mgm.												
None	0.90	0.90	0.90	0.54		0.54	0.31	0.31	0.26	0.60	0.26	0.56	0.56	0.56	0.56
50	1.17	0.76	0.76	4.15		0.54	0.31	0.82	9.35	0.98	0.60	0.56	9.92	2.37	
100	1.17	0.76	0.76	4.69		0.54	0.31	0.82	10.63	1.35	0.60	0.56	9.92	3.73	
300	1.17	0.76	0.76	4.94		0.54	0.31	0.82	9.30	1.50	0.60	0.56	9.92	5.40	
500	1.17	0.76	0.76	5.10		0.54	0.31	0.82	11.42	2.54	0.60	0.56	9.54	6.19	
700	1.17	0.76	0.76	6.70		0.54	0.31	0.82	11.47	1.35	0.60	1.06	10.49	6.63	
900	1.17	0.76	0.76	7.32		0.54	0.31	2.82	11.47	2.12	0.60	2.18	11.32	6.63	
1,200	2.78	0.76	0.76	9.39		0.54	0.31	2.02		3.91	0.60	5.30	11.30	7.35	
1,500	2.78	0.76	0.76			0.54	0.31	2.02	11.77	3.88	0.60	6.20	12.22	6.82	
2,500	3.84	4.92	0.76	11.22		0.54	4.29	4.27	12.94	4.77	0.60	8.50	13.47	16.23	
5,000	7.68	11.76	5.43	11.50		2.37	9.60	8.46	14.76	10.38	3.55	17.74	16.93	16.25	
10,000	16.69	16.87	11.76	16.32		4.74	13.27	10.79	17.76	13.95	6.65	19.48	18.19	16.23	

Series 17. The influence of NaNO₃ upon the accumulation of nitrates formed in the nitrification of various manures

In order to test this out, therefore, a new series of experiments were set up, the extractions from the Carrington loam being used, in which the effect of sodium nitrate had been tested upon the nitrification of cottonseed meal, dried blood and ammonium sulfate (tables 18, 19 and 20). Nitrates were determined colorimetrically by the Greiss method (106). From an analysis of the data in table 23 it will be readily seen, considering the first point raised, that the amount of nitrates formed in any case is not one part per million. This amount would be equivalent to about one drop of N/50 acid, as used in the

reduction method. The figures, then, represent actual quantities of nitrates and not the sum of the two.

To take up next the action of sodium nitrate upon the accumulation of nitrites we find that while there is a general tendency for this radical to increase in amount, it does not reach a concentration of one part per million in any soil until a concentration of from 5,000 to 10,000 pounds per acre is present. At a concentration of 10,000 pounds per acre we find a high amount of nitrite formed in the nitrification of dried blood; on the other hand, only 3.50 and 2.36 parts per million result from the action of sodium nitrate upon the nitrification of the other two materials.

It is clear then that *Nitrobacter* must be influenced in a manner similar to *Nitrococcus*, or else we would get an accumulation of nitrites rather than ni-

TABLE 23

The influence of sodium nitrate on the nitrites formed in the nitrification of various manure

SOIL PORTION	POUNDS PER ACRE NaNO_3	CARRINGTON LOAM	CARRINGTON LOAM	CARRINGTON LOAM
		$(\text{NH}_4)_2\text{SO}_4$	Dried blood	Cottonseed meal
Check	Untreated	0.2143	0.2143	0.2143
1-2	No nitrate + Manure	0.2143	0.7390	0.6420
3-4	50	0.2405	0.7620	0.6630
5-6	100	0.2143	0.9135	0.6940
7-8	300	0.1748	0.9203	0.5570
9-10	500	0.2185	0.9203	0.6995
11-12	900	0.2710	0.8410	0.6630
13-14	1,500	0.2143	0.9500	0.6765
15-16	2,500	0.2185	1.0370	0.7325
17-18	5,000	0.5865	2.3000	0.6200
19-20	1,000	3.5200	10.710	2.3600

trates. The larger accumulation of nitrates at a concentration of 10,000 pounds per acre, would lend support to the opposite view, however. Kelley finds that nitrites are toxic to *Nitrobacter* in large quantities, i.e., 40 mgm. per 100 gm. of soil. Smaller amounts, however, do not affect this condition. As the concentration of sodium nitrate rarely reaches such a figure we need have no fear of nitrites forming in such quantities as to inhibit either plant growth or biological activities.

To summarize the influence of sodium nitrate upon the nitrifying process in the soils it would seem as if the earlier views of Lipman and Brown (109) Crowther (23) and Miyake (127) were substantiated, namely, that sodium nitrate does enhance the nitrification process to a greater or less extent. This stimulating effect may not necessarily be superficially observed, but on the other hand, may cause secondary stimulations to set in, whereby large increases

in the numbers of other soil organisms are brought about, these organisms using the nitrate as well as the nitrifiable material as a source of energy and food. Thus the stimulative effect may be entirely hidden and a decreased accumulation of nitrates be noted instead.

In large quantities; as one would naturally suspect, the presence of nitrates discourages the action of the nitrifying group. In the nitrifying group we have only two known species of bacteria. Necessarily then, we can have no survival of organisms from the action of high salt concentrations, the entire group being eliminated at once.

Deherain (25) says that whereas sodium nitrate may at first depress the activity of the nitrifying group, after some time the organisms readjust themselves to this high concentration and become active. In an experiment carried out by him he added 6 centigrams of sodium nitrate to 100 gm. of fresh soil and noted the nitrates formed at the end of 30 days. He found none; however, shortly after this the soil commenced to nitrify vigorously.

It is also of interest to cite the work of Russel (156) at this point, on the nature and amount of the fluctuation of nitrates in arable soils. This investigator reports that the maximum amount of nitrate accumulation in sand, loams and clays during the year is 6 parts per million in the sands, 14 parts in clays and 23 parts in loams. The action may no doubt also be partially explained by the above phenomenon, viz., the increased cell division, with subsequent transformation of the soluble material.

The fact that nitrates are assimilated again raises the question as to whether or not this does not seriously affect the availability of nitrogenous manures. Being bound up in the body tissue of the microorganisms they must be mineralized during the growing season to be especially beneficial to the crop. If they are not readily simplified a serious problem is at hand, over which the farmer has very little control. The availability of bacterial nitrogen will be studied later on in the text.

In Colorado there are many soils seriously affected by high accumulations of nitrates. Sackett's (147) work would lead one to believe that these nitrate accumulations were of bacterial origin. As the transformation and accumulation of nitrates would have the same scientific interest as the application of sodium nitrate would have upon soils not affected with niter trouble, it was thought that a comparison of the activities of a Colorado "niter spot" soil with the action of sodium nitrate in the above soils would be interesting.

Through Professor Sackett's kindness the author was able to secure such a soil and the data concerning its biological activity are given in series 18.

Series 18. The ammonifying, nitrifying and nitrogen-fixing power of a Colorado niter-spot soil

In collecting this soil, samples were taken under sterile conditions and shipped to the New Jersey Station by express. On arrival they were spread out to dry on a piece of sterile wrapping paper. The soil was a sandy loam

very low in organic matter. All samples were taken from the same field. Sample 1 was taken in the niter spot itself; sample 2 where the effects of the niter spot were just beginning, and sample 3 where no effects were to be noted. The soils were analyzed for total nitrates and chlorides, with the results shown in table 24.

The various biological tests carried out on these samples were ammonification, nitrification and nitrogen fixation. Fixation was carried out in solution because of the lack of soil.

TABLE 24
Nitrates and chlorides in Colorado soil samples

	MILLIGRAM PER 100 GM. OF SOIL	
	NO ₃ as N	Cl
Sample 1.....	4.78	144.87
Sample 2.....	0.86	2.91
Sample 3.....	0.72	1.94

TABLE 25
Ammonifying power of the niter spot samples

SOIL	AMMONIA NITROGEN ACCUMULATION	AVERAGE	PER CENT RELA-TION TO SAMPLE 3
<i>Dried blood</i>			
Sample 1.....	0.93	0.93	0.93
Sample 2.....	18.53	19.45	18.99
Sample 3.....	26.52	26.95	26.73
<i>Cottonseed meal</i>			
Sample 1.....	10.79	10.50	11.14
Sample 2.....	18.15	16.37	17.26
Sample 3.....	23.00	23.00	23.00

The ammonifying power of the soil samples was first studied, dried blood and cottonseed meal being used as the source of organic matter. The data are given in table 25.

It will be seen that, giving sample 3 a value of 100, there is a gradual decrease in the ammonifying power of the samples as the niter spot is approached. Sample 2 has an ammonifying power of 72, on an average, for both sources of organic matter. Sample 1 was not able to ammonify dried blood, although it could simplify cottonseed meal to a large extent. This is in keeping with the increased toxic effect of sodium nitrate where cottonseed meal was used as the source of organic matter in ammonification work. In this case, how-

ever, the decreased activity cannot be due to increases in nitrates, as can be seen from table 23.

The next experiment, recorded in table 25, was carried out to test the nitrifying power of the three samples. By consulting the table we find marked differences in the ability of these three soils to nitrify the three sources of organic matter.

Considering first ammonium sulfate, we find sample 2 nitrifying this material to a very small extent. In fact the nitrification of this substance in sample 2 and that in sample 3 are nearly alike. However, with dried blood a somewhat greater nitrification took place than with ammonium sulfate in samples 2

TABLE 26
The nitrifying power of the niter spot soils

SAMPLE NUMBER	NH ₄ SO ₄			PER CENT RELATION TO CHECK	DRIED BLOOD			PER CENT RELATION TO CHECK	COTTONSEED MEAL			PER CENT RELATION TO CHECK			
	Nitrate accumulation				Nitrate accumulation				Nitrate accumulation						
	M gm. N	M gm. N	Average gm. N		M gm. N	M gm. N	Average gm. N		M gm. N	M gm. N	Average gm. N				
1	1.69	1.79	1.74	16	1.65	1.75	1.70	18	1.57	1.57	1.57	27			
2	1.68	1.68	1.68	16	4.59	4.59	4.72	41	3.93	4.22	4.07	71			
3	10.31	10.33	10.32	100	8.65	9.51	9.08	100	5.90	5.55	5.72	100			

TABLE 27
Nitrogen-fixing powers of the niter spot samples

	NITROGEN FIXED		AVERAGE	PER CENT RELATION TO SAMPLE 3
	M gm.	M gm.		
Sample 1.....	2.47	2.95	2.71	19
Sample 2.....	13.47	11.11	13.29	94
Sample 3.....	14.21	13.97	14.07	100

and 3. Likewise cottonseed meal gave a relative stimulation considerably greater than the other two sources of nitrifiable material. Taking a comparison between the action of sodium nitrate in large quantities upon the nitrification of these nitrifiable materials, we find a close parallelism between the phenomenon there registered and here.

Repeating the work on the nitrogen-fixing powers of these three samples we find from the data in table 27 that there is also a decreased nitrogen fixation as the samples approach the niter spot.

PART IV

Series 19. The influence of sodium nitrate upon nitrogen fixation

The various biological activities already considered deal with the transformation of soil nitrogen. Starting with the complex protein molecule, we have seen how sodium nitrate acts first upon the ammonifying group and second, upon the nitrifying group. Whatever the complexity of these processes, whatever the inter-relation or inter-dependence of the several bacterial factors, they can add nothing to the store of soil nitrogen. Whatever the rate of ammonification, the soil nitrogen cannot thereby increase in amount. Fortunately, there are compensating factors. Just as the nitrogen atoms may be torn apart from those of other elements, so may the latter be made to combine with other elements in the formation of new nitrogen compounds. These compensating factors are the various species of nitrogen-fixing bacteria.

The influence of nitrates as a factor in affecting the activities of the nitrogen-fixing organisms has been studied by a host of investigators, among whom may be cited Heinze (69), Headden (68), Lipman (105), Strank (182), Stocklassa (179) and Sackett (159). So numerous are the experiments upon this phase of the soil nitrogen problem, it is hardly necessary to go into it in any detail other than to corroborate the findings of previous investigators.

In order to throw some added light upon the question of nitrogen-fixation and soluble nitrogen, two experiments were instituted, the one conducted in solution and the other in the Norfolk sandy loam.

The effect of sodium nitrate upon nitrogen-fixation was first studied in solution.

Fifty cubic centimeters of Sohm's soil mannite broth was placed in 500-cc. Florence flasks. Sodium nitrate was then added in quantities as shown in table 28. The flasks were then inoculated with a vigorous culture of nitrogen-fixing organisms.² After inoculation, the flasks were incubated for 21 days at 20°C. At the end of this time the total nitrogen in the system was determined, the nitrates being reduced by means of salicylic acid and zinc dust. The total nitrogen at the beginning of the experiment, as well as the nitrogen in the nitrates added, also was determined.

It will be seen from an observation of the data in table 28, that small amounts of sodium nitrate slightly increased the amount of nitrogen fixed. As the nitrogen increases, however, the fixation rapidly diminishes, depending upon the amount of nitrates in the system. These data seem to be in line with the earlier solution studies of Lipman, Heinze and Stocklassa, i.e., that small amounts of nitrates stimulate the activity of the *Azotobacter*, whereas when larger quantities are present this action is discouraged, the organisms living upon the nitrates.

No work has come to the writer's attention in which this phenomenon has been observed under actual soil conditions. In order to study the process

² The culture was obtained through the kindness of Dr. H. C. Lint, of the Alphano Humus Company, New York City.

in soils, a second experiment was started, the Norfolk sandy loam being used.

To 50 gm. of Norfolk sandy loam in tumblers, were added 2 gm. of dextrose and 2 gm. of the same inoculating material. The whole was then thoroughly mixed together and sodium nitrate added, as is shown in table 28. Water was adjusted to 50 per cent of the water-holding capacity and the soils were incubated for 21 days as before. At the end of this time they were thoroughly air-dried and analyzed for total nitrogen, the nitrates being reduced with salicylic acid and zinc dust.

It will be seen from an examination of table 28 that a similar set of affairs is present in this series as was noted in the solution study, namely, a decrease in the amount of nitrogen fixed varying with the size of the application. At a concentration of 1500 pounds per acre in this experiment, the nitrogen-fixing organisms ceased to fix any more atmospheric nitrogen.

TABLE 28
The effect of sodium nitrate upon the fixation of atmospheric nitrogen

LAB. NO.	NITROGEN FIXATION IN SOLUTION						PER CENT RELATION TO CHECK	NITROGEN FIXATION IN NORFOLK SANDY LOAM						
	NaNO ₃ added	Total nitrogen in solution			Minus no treat- ment	Minus NaNO ₃ added		NaNO ₃ pounds per acre	Nitrogen in 50 grams soil			Minus no treat- ment	Minus NaNO ₃ added	
		Mgm. N	Mgm. N	Average mgm. N					Mgm. N	Mgm. N	Average mgm. N			
1-2	mgm.				mgm.	mgm.						mgm.	mgm.	
1-2	Check	38.14	38.14	38.14				Check	41.87	42.97	42.42			
3-4	None	57.40	56.58	56.99	18.85	18.85	100	None	56.62	55.55	54.08	9.66	9.66	
5-6	5	57.83	57.75	57.99	19.83	19.22	102	100	52.62	54.20	53.41	10.93	9.98	
7-8	10	59.49	59.27	59.38	21.24	19.76	105	200	53.50	53.50	53.50	11.08	8.56	
9-10	25	62.19	61.32	61.75	23.61	19.98	106	600	50.25	51.60	50.92	8.50	4.12	
11-12	50	63.75	63.20	63.47	25.33	17.58	90	1,000	53.35	51.50	52.42	10.00	3.17	
13-14	100	67.42	68.34	67.88	29.74	14.14	75	1,500	50.65	50.65	50.65	8.23	-1.00	
15-16	250		87.25	87.25	49.11	10.37	55						0	

The question is asked whether this discouraged activity is due to a concentration of the sodium salt or to some other cause. We know that sodium nitrate increases bacterial activity to a great extent. Perhaps the presence of this salt increased the bacterial numbers to such an extent that the increased host of microbes used up all of the available energy that the *Azotobacter* require to fix elementary nitrogen. Or again, the increased numbers may have made it difficult for *Azotobacter* to work very handily.

Bearing upon this point Lipman (105) has data showing that *Azotobacter Vinelandii* is seriously inhibited in its own nitrogen-fixing power by the presence of ammonia and nitrate salts.

It would seem, considering the work of the earlier investigators as well as the experiments tabulated here, that the nitrogen-fixing power in the soil is discouraged by applications of sodium nitrate, although in solution a slight

stimulating action is to be seen. This decreased fixation is to be explained on several grounds, among which are concentration, antagonism of other organisms, the using up of the energy supply by other organisms, and the discouragement of fixation by the use of sodium nitrate as a food by *Azotobacter*.

PART V

Series 20. The transformation of nitrates by soil microorganisms

Experiment 1. It has been shown in the previous pages that soil micro-organisms are able to use soluble nitrogen in their life processes. Moreover, it has developed that the amount and efficiency with which they assimilate ammonia or nitrates depends to a very large extent upon the kind and amount of energy-providing material. All other things being equal, the assimilation is in direct proportion to the supply of energy.

Necessarily, soils must vary in the amounts of available energy which they contain. The great mass of organic matter left in the roots and that added annually to the soil in the form of green and farm manures contains a low percentage of nitrogen and a high proportion of energy-providing materials. This excess energy must affect the activity of the soil organisms, and hand and hand with this the soluble plant-food in soils.

Again experiments carried out in solution have indicated that the form in which the various nitrates are supplied has much to do with their assimilation. Kossowitz (92) and de Grazia (53) have published data to the effect that calcium nitrate is more resistant to transformation than sodium nitrate.

In order to throw some light upon this point with regard to the same action in soils a new series of experiments were set up to test the relative assimilability of sodium nitrate, potassium nitrate, and calcium nitrate in three types of soil; also the relative assimilability of these nitrates when supplied to a soil with large amounts of energy material.

The experiments were carried out as follows:

One-hundred-gram portions of three types of soil, the Carrington loam, Norfolk sandy loam, and the Penn clay loam were placed in tumblers and the three nitrate-bearing salts added to them in amounts equal to 200 mgm. of the salt. Water was then supplied to 50 per cent of the water-holding capacity. The tumblers were covered with glass plates and incubated at 20°C. for 21 days. Where organic matter was supplied this was added to the soil by means of the mixer previously cited. At the beginning and at the end of the experiment the soils were extracted with water until they showed the presence of no nitrates by the diphenylamine reaction as outlined by Withers and Ray (224).

It is assumed that the soluble nitrogen not in the form of ammonia or nitrates has been attacked by microorganisms in such a manner as to make them of no use for plant growth.

The data recording the transformation of the three nitrate-bearing salts in the three above-mentioned soils are given in table 29. In this table columns 1 and 2 express the ammonia present in the soils at the beginning and at the

end of the experiment, column 4 the amount of nitrate applied in milligrams of nitrogen, column 3 the total nitrates in the soil including the soil nitrates, column 5 the total nitrates at the end of the experiment, column 6 the total applied nitrates at the end of the experiment, column 7 the milligrams of total applied nitrates transformed and column 8 the per cent of applied nitrogen transformed.

From an examination of the table it is to be seen that the total amount of nitrates assimilated varies from 15 to 30 per cent with a general average of about 22 per cent in all soils and with all sources of nitrates. In the Norfolk sandy loam 23.56 per cent of the nitrogen of sodium nitrate was transformed, in the Penn loam clay 20 per cent and in the Carrington loam 24.27

TABLE 29
Transformation of $Ca(NO_3)_2$, KNO_3 , and $NaNO_3$ in various soils

LAB. NO.	SOIL AND TREATMENT	NH ₃ AT BEGINNING MGM. N	NH ₃ AT END MGM. N	TOTAL NO ₃ AT BEGIN- NING MGM. N	N AS NO ₃ APPLIED MGM. N	TOTAL NO ₃ AT END MGM. N	TOTAL APPLIED NO ₃ AT END MGM. N	APPLIED NITROGEN TRANSFORMED MGM. N	PER CENT OF APPLIED N TRANSFORMED
1-2	N. S. L. no treatment.....	0.42	0.45	1.67	.	3.01			
3-4	N. S. L. + NaNO ₃	0.42	0.48	30.02	28.35	24.68	21.67	6.68	23.56
5-6	N. S. L. + KNO ₃	0.42	0.47	39.17	37.50	31.79	28.78	8.72	23.26
7-8	N. S. L. + Ca(NO ₃) ₂	0.42	0.45	34.42	32.75	30.67	27.66	5.09	15.52
9-10	P. C. L. no treatment.....	0.28	0.28	2.25		7.89			
11-12	P. C. L. + NaNO ₃	0.28	0.29	30.60	28.35	30.57	22.68	5.67	20.00
13-14	P. C. L. + KNO ₃	0.28	0.28	39.75	37.50	37.08	29.19	7.31	19.45
15-16	P. C. L. + Ca(NO ₃) ₂	0.28	0.28	35.00	32.75	31.19	23.30	9.45	28.85
17-18	C. L. no treatment.....	0.27	0.27	3.37		6.38			
19-20	C. L. + NaNO ₃	0.26	0.28	31.72	28.35	28.84	21.47	6.88	24.27
21-22	C. L. + KNO ₃	0.28	0.27	41.87	37.50	36.00	29.63	7.87	20.99
23-24	C. L. + Ca(NO ₃) ₂	0.28	0.28	36.12	32.75	31.43	25.06	7.69	23.45

per cent. The amount of nitrogen as calcium nitrate assimilated seems to be at a minimum in the Norfolk sandy loam and at a maximum in the Penn clay loam. Potassium nitrate was effectively transformed in the Carrington loam and Penn clay loam soils.

Comparing the assimilation of calcium nitrate with that of the other two salts it can not be said, as a generalization, that it is more resistant to transformation than they are. The relatively large percentage of assimilation of the three salts is quite remarkable and may well enter into the availability question as a partial explanation of the lack of recovery of the total nitrogen supplied in mineral fertilizers. In these three soils the different amounts of organic matter no doubt supplied the energy for the above phenomenon. We can, however, cause a similar phenomenon to take place by using standard soils and supplying organic matter in various forms.

Experiment 2. In this experiment the effect of barley straw was studied on the assimilation of these three salts. It will be seen by consulting table 30 which is constructed on the same order as table 29 that very much larger amounts of soluble nitrates are taken out of solution. The greatest amount of nitrate assimilated in this experiment was in the form of potassium nitrate. As was the case with this soil in the previous experiment, calcium nitrate was the least assimilated.

The phenomenon of assimilation has been predominant in all of the previous series of investigations. Once in the protoplasm of the microorganisms it must again become mineralized before it can be used by the plant. It is of great importance to us to know how fast this combined nitrogen will become available again. Beijernick (7) has some data on this point. He found that some 50 per cent of the total nitrogen of *Azotobacter* cells which he supplied was

TABLE 30

The transformation of nitrates from various sources in a Norfolk sandy loam rich in organic matter

SOIL PORTION	SPECIAL TREATMENT	NH ₃ AT BEGINNING MGM. N	NH ₃ AT END MGM. N	APPLIED NO ₃ AT BEG- GIVING MGM. N	TOTAL NO ₃ AT BEGIN- NING MGM. N	TOTAL NO ₃ AT END MGM. N	SOIL NITRATES TRANS- FORMED MGM. N	APPLIED NO ₃ AT END MGM. N	APPLIED NO ₃ TRANS- FORMED MGM. N	PER CENT APPLIED NO ₃ TRANSFORMED	PER CENT OF SOIL NO ₃ TRANSFORMED
1-2	No treatment.....	0.42	0.45		1.67	3.01					
3-4	2 per cent Barley straw.....	0.42	0.28		1.67	1.09	1.92				63.78
5-6	2 per cent Barley straw + NaNO ₃	0.42	1.65	28.35	30.02	14.22		13.13	15.22	53.69	
7-8	2 per cent Barley straw + KNO ₃	0.42	1.67	37.50	39.17	14.79		13.70	23.80	63.48	
9-10	2 per cent Barley straw Ca(NO ₃) ₂	0.42	1.69	32.75	34.42	20.00		18.91	13.84	42.27	

nitrified in about seven weeks' time. Biereima (10) also has worked on the question. He found that the condition of the protoplasm was a limiting factor in the process of mineralization. Organisms having a large abundance of spores nitrified very little, whereas those not in this condition were nitrified some 20 to 40 per cent in two months. The writer has also carried out an experiment on this point in which the nitrifiability of some common soil organisms has been studied. The method of obtaining the microorganic substance is as follows:

A sterile portion of Cook's (19) No. 2 medium was inoculated with pure cultures of soil fungi and incubated for 12 days. At the end of this time the microbial matter was killed by sterilization and the dead substance filtered off upon filter paper and washed free from nitrates. The material was then dried at 100°C. and ground with nitrogen-free quartz sand.

A similar process was used to obtain a bulk of *B. mycoides* protoplasm. In this case, however, sterile bouillon was inoculated with this organism and at the end of the incubation period the bacterial mass was removed by means of a Berkefeld filter, washing the substance with distilled water until free from foreign matter. It was then dried and ground with sand. A culture of *Actinomyces penicilloides* was also obtained on the same manner as the fungus material was secured.

Total nitrogen determinations were then made upon the sand plus microorganisms and enough of the material to equal 21 mgm. of nitrogen was used to test its nitrifiability. The Carrington loam was used, as this soil had a powerful nitrifying flora.

For comparative purposes the nitrifiability of cottonseed meal, alfalfa, dried blood and green rye were likewise studied at the same time.

In recording the data given in table 31 the relative values of the materials have been worked out, giving dried blood a value of 100. It is readily seen from a survey of table 31 that there is quite a variation in the nitrifiability of

TABLE 31
Nitrification of microorganic substance

SOURCE OF NITROGEN	NO ₃ ACCUMULATION		AVERAGE MGM. N	PER CENT RELATION TO DRIED BLOOD
	MGM. N	MGM. N		
<i>Actinomyces penicilloides</i>	0.18		0.18	0
<i>Alfalfa meal</i>	6.10	2.77	6.43	41.0
<i>Aspergillus niger</i>	6.20	6.30	6.25	41.0
<i>Bacillus mycoides</i>	1.18	1.18	1.18	0.5
<i>Cottonseed meal</i>	9.86	9.96	9.91	65.0
<i>Cladosporium herbarum</i>	4.42	4.42	4.42	28.0
<i>Dried blood</i>	15.23	15.39	15.31	100.0
<i>Fusarium bullatum</i>	15.39	9.03	12.21	79.0
<i>Mucor spinosus</i>	9.93	7.24	8.58	76.0
<i>Rhizopus tritica</i>	10.63	8.93	9.78	65.0
<i>Rye</i>	2.20	2.05	2.12	1.0

microbial material, the nitrates formed varying from 0 to 79 per cent of the value of the dried blood.

The organisms most readily nitrified were *Fusarium bullatum* and *Mucor spinosus*. The two organisms were even more nitrifiable than an equivalent quantity of nitrogen as cottonseed meal or alfalfa meal. *Rhizopus tritica* also nitrified as well as cottonseed meal, and superior to rye or alfalfa. In fact, rye did not nitrify at all during the incubation period. *Aspergillus niger*, very rich in spores, was of low nitrifiability; this was likewise true of *Cladosporium herbarium*.

The only bacterium and *Actinomyces* experimented with nitrified but little. Bierima's (10) data on this point, however, give the nitrifiability of *Bacillus agrestis* as 18 per cent, *Bacillus radiobacter* 9 per cent and *Bacillus fluorescens* 18 per cent of the total nitrogen supplied, at the end of 2 months.

Taking the whole information on this subject *per se*, it would appear that this nitrogen would probably not become available for crop use until too

late in the season. This would again strengthen our stand that the assimilation of soluble mineral fertilizers has much to do with the ultimate recovery of applied nitrogen.

In the preceding pages an attempt has been made to study the influence of sodium nitrate upon the nitrogen transformation in soils, with a view of obtaining a more perfect understanding of the various factors which go to affect the availability of this manure. Some of the more pertinent points brought out in the investigation have been summarized below. Necessarily all the details cannot be summarized and the reader must consult the text for the finer points of the investigation.

In carrying out the work the author has had the ever helpful advice of Dr. J. G. Lipman, whom he now takes the opportunity to thank most heartily. In addition the author wishes to thank Prof. A. W. Blair, Dr. William S. Myers and the members of the Soil Fertility Research Laboratory of Rutgers College for their kindly interest and suggestions during the course of the investigation.

SUMMARY

1. It has been shown by a review of the literature upon the availability of nitrogenous manures that a given quantity of nitrogen in the form of nitrates is superior to the same quantity in the form of ammonia and this in turn is of more value than organic forms.
2. Theories for these relative availabilities have been presented and discussed.
3. The reasons why a higher proportion of the applied nitrogen of sodium nitrate is not recovered are also discussed and the probability of these differences being explained on biological grounds is studied in detail.
4. In studying the influence of sodium nitrate upon nitrogen transformation in soils, its effect upon the ammonifying, nitrifying and nitrogen-fixing powers of seven types of soil has been taken up.
5. It has been shown that applications of sodium nitrate markedly increase the simplification of protein material applied to soils.
6. There is some difference to be noted, however, with regard to the source of organic matter.
7. Cottonseed meal is ammonified to a much larger extent in the presence of sodium nitrate than is dried blood.
8. The lesser effect of the action of sodium nitrate upon the ammonification of dried blood is only apparent, and not real.
9. This is due to the effect of this fertilizer in increasing bacterial activity, these increased numbers assimilating the end point.
10. A method for studying this assimilating phenomenon has been outlined and successfully used.
11. Acid phosphate increases the ammonification of dried blood nitrogen, but sodium nitrate added as a limiting factor does not enhance the decay of this material.

12. Potash in the form of potassium chloride has a slight stimulating effect in some soils. In others, however, no action is to be observed.
13. Sodium nitrate decreases the ammonia accumulation in soils supplied with excess energy in the form of dextrose.
14. If arranged in the proper proportions, sodium nitrate, acid phosphate and potassium chloride markedly increase the simplification of organic matter, in a degree beyond which any single application would stimulate.
15. Sodium nitrate loses its stimulating action to a great extent in alkaline soils.
16. This is due to an increase in the numbers of bacteria, which assimilate a considerable proportion of the simplified material, and also to a rearrangement of the soil flora.
17. Of the soil flora studied the soil fungi respond the most to applications of sodium nitrate, with the bacteria next in order.
18. The stimulating influence of sodium nitrate is due to the anion.
19. Sodium nitrate stimulates the nitrification of dried blood, cottonseed meal and to a less extent ammonium sulfate.
20. The stimulative action is not apparent. Secondary reactions, such as increased cell division, with a subsequent assimilation of nitrates, are set up in the system and hide the end point.
21. In large quantities sodium nitrate depresses nitrification, the magnitude of the depression depending first upon the sources of nitrifiable material and second upon the type of soil.
22. In large quantities the presence of sodium nitrate first becomes toxic to the nitrification of ammonium sulfate, then to dried blood and lastly to cottonseed meal.
23. Sodium nitrate in amounts up to 5000 pounds per acre affects *Nitrobacter* the same way as *Nitrococcus*.
24. In amounts beyond 5000 pounds per acre there is evidence to show that *Nitrobacter*'s activity is stopped, whereas, that of *Nitrococcus* still manifests itself.
25. Sodium nitrate in small quantities stimulates nitrogen fixation by *Azotobacter*. In larger quantities this salt depresses this process.
26. Large quantities of nitrates are assimilated by biological forms in soils.
27. The amount assimilated is about 20 per cent of the applied nitrogen.
28. The idea is not tenable that calcium nitrate is assimilated to a lesser degree than sodium nitrate.
29. Experiments carried out on the nitrifiability of microbial matter show wide differences in this respect.
30. The entire study of the influence of the sodium nitrate, upon nitrogen transformations in soils seems to indicate rather strongly that in the cases where larger quantities of nitrogen are recovered in the crop than can be accounted for by the amount of sodium nitrate applied, this is due to a drawing

on the soil's own nitrogen supply. This supply is acted upon by a stimulated bacterial flora, brought about by the presence of sodium nitrate.

31. On the other hand, where more or less of the nitrogen applied is recovered the variations in the recovery may in a large measure be explained on the grounds of assimilation of nitrates by soil organisms.

32. Of the three nitrogen-transforming groups of soil organisms sodium nitrate affects the nitrogen-fixing group most adversely, has a lesser detrimental effect on the nitrifying group, and affects the ammonifying group least adversely.

33. As applied in agricultural practice sodium nitrate generally enhances the activity of the ammonifying and nitrifying groups. On the other hand, the activity of the nitrogen-fixing group is discouraged by its presence.

34. In no case will toxicity be caused by sodium nitrate if it is applied rationally.

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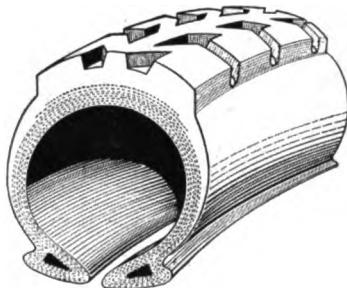
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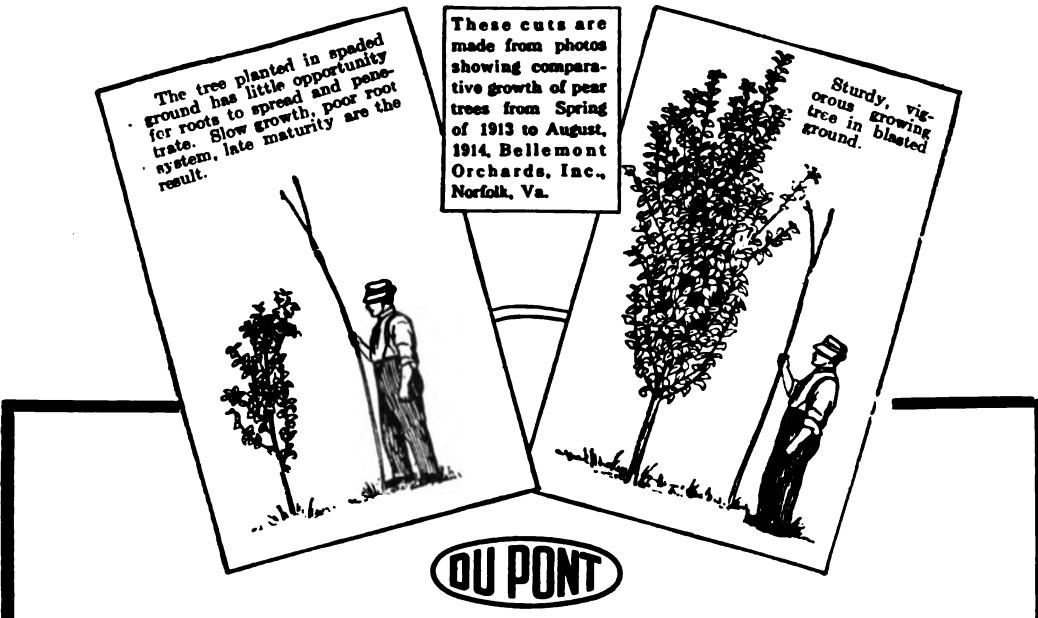
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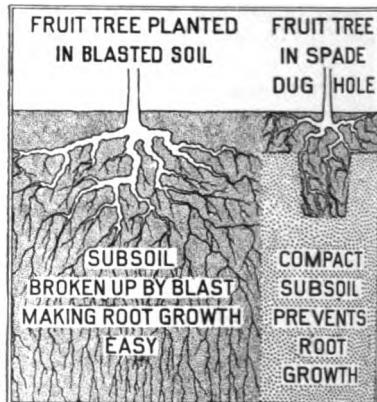
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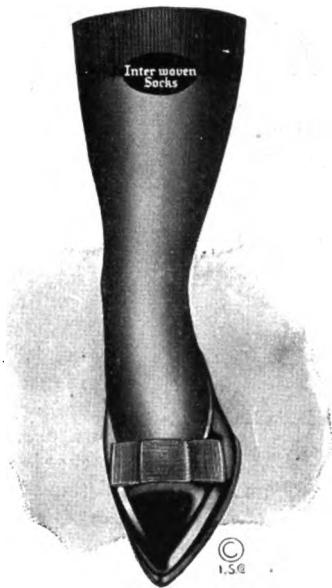
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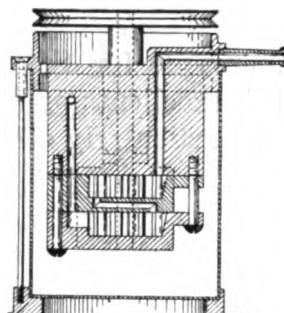
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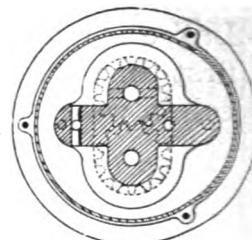
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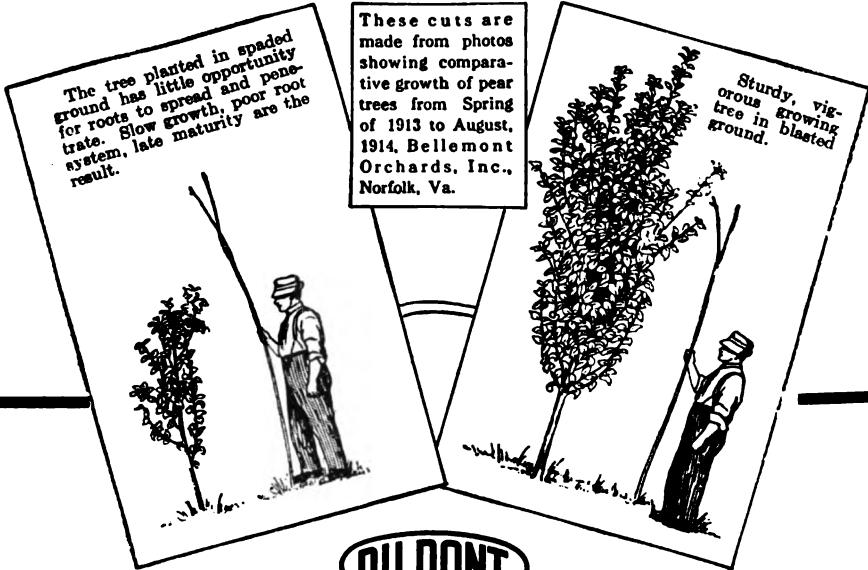
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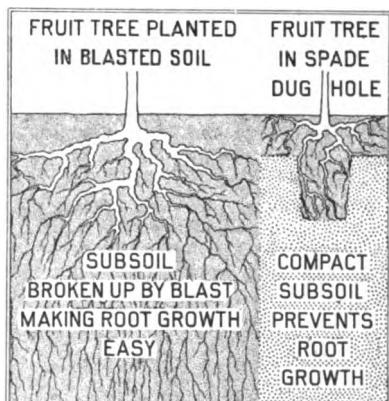
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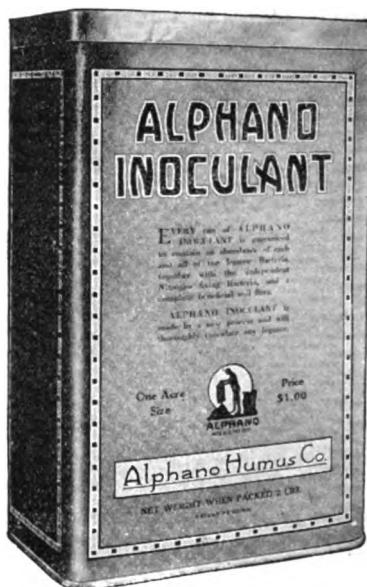
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A CORRELATION BETWEEN BACTERIAL ACTIVITY AND LIME REQUIREMENT OF SOILS

FIRMAN E. BEAR

Department of Agricultural Chemistry and Soils, Ohio State University

Received for publication September 12, 1917

INTRODUCTION

Limestone regions are noted for their fertility. Alfalfa, red clover, blue-grass, and corn are among the crops which thrive best on limestone soils. Those soils which do not naturally contain carbonate of lime are usually made more productive by applications of lime or limestone. Extensive investigations carried out by the Rhode Island, Maryland, Pennsylvania, Ohio, Illinois, and other agricultural experiment stations have demonstrated the value of lime in either the oxide, hydrate or carbonate form on soils which are acid to litmus. An excellent review of the most important investigational work on the use of lime on acid soils is given by Frear (9).

The investigations of Wheeler at the Rhode Island Agricultural Experiment Station, indicate, however, that a number of plants of economic importance thrive on soils which contain no solid carbonate of lime. Some of these plants are benefited by lime, but others are injured by applications of lime. Wheeler (36) says that orchard grass (*Dactylis glomerata*, L.) and meadow fescue (*Festuca elatior*, L.) are less injured by soil acidity than Kentucky blue-grass (*Poa pratensis*, L.) and timothy (*Phleum pratense*, L.) and that awnless brome grass (*Bromus inermis*, L.), red top (*Agrostis alba* var. *vulgaris*, Thurb.), and Rhode Island bent (*Agrostis canina*, L.) do not seem to be susceptible to injury even on decidedly acid soils. He also states (37) that Concord grapes are apparently indifferent to the lack of lime and that cranberries, raspberries, and lima beans are injured by liming, the last named growing splendidly on soils so acid as to entirely destroy lettuce, spinach, onions, beets and asparagus. In his latest publication on this subject Wheeler (38) gives a summary of his work in which he shows that plants vary in their requirements from those which are injured by applications of lime even to a very acid soil, to those which are unable to live on an acid soil and are benefited remarkably by lime.

Coville (6) states that the blueberry, cranberry, strawberry, blackberry, red raspberry, potato, sweet potato, rye, oats, millet, buckwheat, red top, carrot, turnip, cowpea, hairy vetch, crimson clover, soybean, lupine, and serratula are adapted to acid soils. He concludes, "soil acidity is not always

an objectionable condition which invariably requires lime" and "under certain conditions, a complete system of acid agriculture is practicable."

Harter (14) writes that liming has been shown to be beneficial to all crops on Norfolk soils with the exception of beans, peas, and tomatoes. Kossovitch and Althausen (26) report that, while the liming of acid podzol soils strikingly increases the yields, the limit of increase is at about the point of neutralization and that an excess injures the plants. No statement is made as to how the point of neutralization was determined. Heinrich (15) concludes that the determination of lime in a soil, by digesting with 10 per cent hydrochloric acid, can be used as an index in determining what crops will thrive. According to his report, the least amounts of lime which will permit of successful growth are:

Crops	Calcium carbonate in the soil per cent
Lupines, potatoes, and rye.....	0.05
Oats and barley.....	0.05 to 0.10
Peas and vetch.....	0.10
Red clover.....	0.10 to 0.12
Alfalfa.....	0.20 to 0.30

Fred and Graul (10) experimenting with alfalfa, soybeans, and red clover on acid soils of two series, conclude that half enough lime to neutralize the soil acidity as measured by the Truog (32) method is sufficient for the production of good yields of these crops on acid soils of these two series.

THE RELATION BETWEEN BACTERIAL ACTIVITY AND THE REACTION OF SOILS

One of the reasons usually given for the maintenance of a neutral or slightly alkaline reaction in soils is that the soil microorganisms, which have to do with the processes of decay and the changes by which certain organic and inorganic substances become available for higher plants are unable to work to best advantage in an acid medium. The ammonifying, nitrifying, and nitrogen-fixing bacteria are thought to prefer a neutral or slightly alkaline medium. However, it is probably true that the various groups of soil bacteria are differently affected by the soil reaction. The influence of acidity and alkalinity on the development of pathogenic bacteria has been studied by a number of investigators. The literature on this subject is reviewed quite fully by Itano (21). The degree of acidity or alkalinity which the organisms are able to withstand varies with the species. Certain forms, e.g., *Bacterium tuberculosis*, are able to live in the presence of a considerable degree of acidity. It is reasonable to believe that soil microorganisms show similar differences in this respect. The fact that many acid soils are supporting vegetation, indicates that bacterial processes are being carried on in them, although these processes might be materially hastened if lime were applied.

The number of bacterial colonies from soil aliquots which will develop on

agar plates is influenced by the reaction of the medium. Hoffmann (16) finds in counting the number of bacteria in soils that a medium slightly acid to phenolphthalein is more favorable than a medium which is neutral or slightly alkaline to phenolphthalein. Fischer (8), who conducted probably the most extensive investigations on the effect of lime on the number of bacteria in soils, shows that an application of either calcium oxide or calcium carbonate has a very marked effect in increasing the total number of bacteria.

That the rate of ammonification is increased by applications of lime is shown by Voorhees and Lipman (35). Coville (6) points out that many soils acid to litmus contain large amounts of ammonia. Kopeloff (25) shows that "where the soil reaction is unfavorable for the activities of the soil bacteria concerned in ammonification, the soil fungi may prove to be an important compensating factor."

The rate of nitrification is increased by applications of lime on soils which give an acid reaction with litmus. The results obtained by Lyon and Bizzell (27) are typical. A number of other investigators report similar effects from the use of lime. Scales (29), studying the activities of nitrifying organisms, finds they are most active in the presence of 50 per cent of the calcium-carbonate requirement (Veitch) of the soil. An excess of calcium carbonate seems to be toxic to the nitrifying organisms. Temple (31) finds that if an organic source of nitrogen is used instead of ammonium sulfate, the formation of nitrates is much greater in acid soils. He explains this increased nitrification on the basis of the formation of neutral zones, caused by the production of ammonia, at which points conditions are favorable for nitrification. Temple also shows that calcium salts of organic acids can be used as effectively as calcium carbonate in overcoming the toxic effect of ammonium sulfate on an acid soil. Miller (28), working with a sandy soil acid to litmus, finds that an application of 0.1 per cent of calcium oxide caused a decrease in the ability of the soil to nitrify ammonium sulfate and that 0.5 per cent of calcium oxide stopped the process entirely. Hutchinson (19) finds that calcium oxide acts not alone as a neutralizing agent, but also as a partial sterilizing agent. Since in the experimental work following applications of neutralizing agents are confined to calcium carbonate, it does not seem necessary to include any further discussion on the effect of calcium oxide on the bacterial processes in the soil.

It should be remembered that it has been shown that nitrate nitrogen is not necessary for all plants. Hall and Miller (12) call attention to the fact that ammonium sulfate, on the Park plats of the Rothamsted Farm, produces very good crops of grass, although the soil is deficient in lime and very little nitrification takes place. Hutchinson and Miller (20) find that peas are able to utilize ammonia nitrogen as well as nitrate nitrogen, although the opposite is true with wheat. Kelley (24) shows that rice, grown in swamp land, secures its nitrogen in the form of ammonia. If ammonification processes are less affected than nitrification processes by a deficiency of lime in

the soil, then plants which are able to utilize ammonia can survive where those depending on nitrate nitrogen cannot live.

Hopkins (18) notes that the application of lime increases the power of *Bacillus radicicola* in certain legumes to fix atmospheric nitrogen. Whiting (39) writes that nodules are often found in abundance on legumes on very acid soils. Japanese clover (*Lespedeza*) has often been observed by the writer growing on soils strongly acid to litmus and the roots were well supplied with nodules. These nodules were mostly near the surface of the soil. Kellerman and Robinson (22) find that crimson clover inoculation is little affected by the reaction of the soil. Fred and Graul (10) find that, if acid Colby silt loam soil is previously inoculated with *B. radicicola*, nitrogen fixation by soybeans is little influenced by applications of calcium carbonate. They also find this true on acid Colby silt loam with red clover. Both clover and alfalfa were able to fix considerable amounts of nitrogen when growing on Colby silt loam and Plainfield sand having only one-half of their acidity (Truog method) neutralized. The Colby silt loam required 10,400 and the Plainfield sand 5200 pounds of calcium carbonate to neutralize one-half of the acidity in 2,000,000 pounds of soil. Determinations of the lime requirement (Veitch) on the Colby silt loam soil, chosen from the same locality the year previous, showed a need of 3234 pounds of calcium carbonate per 2,000,000 pounds of soil. The authors state that "the Truog method shows much larger amounts of soil acidity than the Veitch."

Ashby (1) shows that the use of lime on the Rothamsted soils more than doubled the nitrogen-fixing power of the *Azotobacter*. Hoffman and Hammer (17) find that calcium carbonate is essential to non-symbiotic nitrogen fixation, but that the amount required is very minute and was present in sufficient amount in all the soils tested. These soils were chosen from various localities in Wisconsin and must have included some soils acid to litmus, since Whitson and Weir (40) estimate that two-thirds of the soils of Wisconsin are acid. Christensen and Larsen (4) find that if Ashby's solution is inoculated with a soil in need of lime, the brownish film usually produced by *Azotobacter* does not develop. They suggest this as a method of determining the need of a soil for lime.

Gimingham (11) describes several organisms capable of bringing about the formation of carbonates from calcium salts of organic acids. Hall and Miller (13) also report that calcium salts of organic acids are transformed to the carbonate by soil organisms, the organic acids being decomposed to form carbon dioxide and water. Drew (7) shows that marine bacteria precipitate calcium carbonate from sea water. He names the organism responsible for this reaction, *Bacillus calcis*. Kellerman and Smith (23) write that it is possible in the laboratory to produce calcium carbonate by three types of biological processes; by the action of ammonium carbonate on calcium sulfate; by the action of ammonium hydroxide on calcium acid carbonate, and by the decomposition of calcium salts of organic acids. They state that Drew's organ-

ism is *Pseudomonas calcis*. This is a denitrifying organism which produces ammonia by the reduction of nitrates. Bear and Salter (2) show that the lime requirement (Veitch) of the West Virginia Agricultural Experiment Station fertility plots is less where the content of organic matter has been increased, and suggest that this decrease may have been due to the precipitation of calcium from solution by the humus in the soil, whereby it was prevented from being lost in the drainage water. This calcium might later be freed as the carbonate, as the decomposition of the organic matter was brought about by the soil organisms.

OBJECT OF THESE INVESTIGATIONS

In view of the fact that large areas of land are acid and that the distance from the supply of lime often makes the cost of applying large amounts of lime or limestone prohibitive, it was thought it might be desirable to consider more carefully the possibilities of a system of acid agriculture as suggested by Coville (6). Since the problem of the economy of nitrogen and its availability for the use of crops is largely a bacterial problem, it seemed important to study the relation of the reaction of the soil to the activities of the bacteria concerned in nitrogen accumulation and transformations. Recognizing the fact that plants do grow on soils which are acid to litmus, how are these plants supplied with nitrogen? We know that lime and limestone are valuable soil amendments, but might it not be possible that small applications of these materials would be relatively more effective in promoting the activities of the bacteria concerned in the nitrogen problem than large applications? If the *B. radicicola* of some legumes is more resistant to acidity than the *B. radicicola* growing on other legumes, might it not be possible to select legumes adapted to the reaction of the soil instead of adding lime to the soil to make the reaction suitable for the legumes we desire to grow? Even if nitrogen-fixing organisms are able to grow in acid soils, are they able to fix atmospheric nitrogen in such an environment? To answer these questions, it was proposed to measure the activities of those bacteria concerned in the nitrogen economy of plants as influenced by various amounts of calcium carbonate applied to acid soils.

DEFINITION OF "LIME REQUIREMENT"

In the preceding discussion, a rather loose construction is given to the term "soil acidity." This is simply in accordance with precedents set by the various investigators whose work is reviewed. As a rule, an "acid" soil means a soil which changes blue litmus paper red. The "degree of acidity" of soils has no such definite meaning, consequently the investigations reported are not strictly comparable. The writer sees no reason to disagree with Truog (33) as to what "soil acidity" really is. Truog writes that acid silicates are the main cause of soil acidity in upland soils. His excellent review of this subject gives a select bibliography of the investigational work along this

line. Truog (32) also writes that the acidity of soils may be conveniently divided into two classes, "active" and "latent" acidity. He states that "latent" acidity is undoubtedly much less injurious to plants than "active" acidity. He also shows the desirability of knowing the "avidity" of the active soil acids. Sharp and Hoagland (30) attempt to measure the lime requirement of soils by determining the hydrogen-ion concentration of the soil suspensions. The recent review of Clark and Lubs (5) of the literature on this subject, indicates that the hydrogen-ion concentration of the medium is the important factor to consider in the relationship between acidity and biological processes. The hydrogen-ion concentration of a soil in suspension in water is, however, not a measure of the amount of lime necessary to add to an acid soil to produce a neutral reaction of the soil. This is partly because of the slow solubility of the acid-forming constituents present in soils.

At the time this investigation was begun, most of the recent work on soil acidity had not been published. The writer felt at that time that the most satisfactory measure of the "lime requirement" of a soil was that obtained by the Veitch (34) method. Accordingly, this method was used in determining the quantitative need of the soils used for lime. It is interesting to note in this connection that when the two soils which were used most largely in these investigations had been treated with the quantity of calcium carbonate necessary to satisfy their lime requirements (Veitch) and had been mixed once each week for 12 weeks, they were found to be neutral to litmus paper.

HISTORY OF THE SOILS USED IN THESE EXPERIMENTS

A large part of the work reported has been done on samples of soil from two different localities belonging to different soil series. Both of these were acid in reaction, as will be shown later.

Soil I was secured from plot 18 of the West Virginia Agricultural Experiment Station farm. The soil is classified by the United States Bureau of Soils as Dekalb silt loam. It is a residual soil which has been formed by the disintegration of sandstone and greenish gray shales overlying the Pittsburg coal. The original timber was largely oak and chestnut with an occasional locust. The analysis of this soil is as follows:

Element	<i>Pounds per 2,000,000 of soil</i>
Nitrogen.....	1,940
Phosphorus.....	600
Potassium.....	25,100
Carbon.....	23,900
Calcium.....	2,300
Magnesium.....	4,300
Calcium carbonate requirement (Veitch).....	3,500

Plot 18 has not received any fertilizer, lime or manure since the beginning of the fertilizer tests in 1900. Only a partial record of the produce of this

plot is available. During a part of the time since 1900 a tile drain, which passed near this plot, was not working, and, since the yields of the plot were somewhat abnormal, no permanent records of the plot were kept. Later the record of the produce of this plot was continued. This record shows that plot 18 corresponds normally in productivity to plot 21, which also received no fertilizer, lime or manure. The sample of soil was chosen from plot 18 because its record was incomplete and any change due to the removal of a large sample of soil would not interfere with the plot experiments. Since 1900 the following crops have been grown on these plots; rye, 1900 and 1907; wheat, 1901 and 1914; clover, 1902, 1909, and 1915; corn, 1903, 1905, and 1912; cowpeas, 1904; potatoes, 1906; timothy, 1909, 1910, and 1911, and oats, 1913. Table 1 gives the records of the fertilizer treatment and total produce of all the plots up to and including 1915.

TABLE 1

Total amounts of fertilizers applied and total produce per acre from 1900 to 1915 on soil I

PLOT	TREATMENT	NITRATE OF SODA pounds	ACID PHOSPHATE pounds	SULFATE OF POTASH pounds	LIME (CaO) pounds	MANURE tons	TOTAL PRODUCE pounds
19	N, P, K, CaO.....	4200	4200	1625	4500		120,605
20	M, CaO.....				4500	210	152,400
21	Check.....						38,600
22	CaO.....				5500		36,615
23	Ash M, N.....	300			Ash of 40 tons of manure until 1912		39,270
24	Check.....						43,075
25	M.....					190	139,670
26	N, P, K.....	4200	4200	1625			117,910
27	Check.....						42,170
28	P, K.....		4200	1625			76,995
29	N.....	4200		1625			52,215
30	Check.....						39,480
31	N, P.....	4200	4200				95,940
32	K.....			1625			41,565
33	Check.....						36,845
34	P.....		4200				63,415
35	N.....	4200					41,195

N, indicates nitrate of soda; P, acid phosphate; K, sulfate of potash; M, manure.

Soil II was secured from the Ohio Agricultural Experiment Station farm at Wooster. This soil is classified by the Bureau of Soils as Wooster silt loam. It has been formed from the disintegration of sandstone and shales of the Mississippian period, under the influence of glacial action. The analysis of the soil used is as follows:

Element	Pounds per 2,000,000 of soil
Nitrogen.....	1,775
Phosphorus.....	664
Potassium.....	34,000
Carbon.....	22,200
Calcium.....	4,470
Magnesium.....	6,596
Calcium carbonate requirement (Veitch).....	3,500

It will be observed that soil II has the same calcium-carbonate requirement as soil I.

Soil II has never received any fertilizer, lime or manure since the beginning of the fertilizer tests in 1893. Continuous records since that time have been kept on soil of the same history as this soil in a 5-year rotation experiment at the Wooster station. The rotation has been corn, oats, wheat, clover, and timothy. A summary of the effect of lime and fertilizers on this soil is given by Williams (41) in table 2. An experiment has also been in progress on this same type of soil which had been kept in a fair state of fertility by a good rotation and an occasional application of manure previous to the beginning of the experiment. The rotation since practiced has been corn, oats, and clover. The records of this experiment are shown in table 3. It will be seen by a study of tables 2 and 3, that both lime and acid phosphate are very effective in increasing the yields of the crops grown in these two rotations. While lime is very efficient, it seems remarkable that such large yields of these crops can be produced by the use of acid phosphate alone on a soil which has a calcium-carbonate requirement of 3500 pounds per 2,000,000 pounds of soil.

The other samples of soil used in these experiments were Dekalb soils chosen from various localities in West Virginia. These soils vary greatly because of differences in the systems of management they have undergone. Analyses of these soils are shown in subsequent tables.

PLAN OF THESE EXPERIMENTS

Large samples of soils, acid to litmus, were secured, sent immediately to the laboratory, made to pass a 2-mm. sieve, and stored in large galvanized iron cans. From these cans soil was removed as needed. Careful analyses of the soils were made for the total amount of nitrogen, phosphorus, potassium, calcium, magnesium, and carbon. Lime-requirement determinations were made by the Veitch method as indicated above. Amounts of C. P. calcium carbonate varying from 250 pounds to 40,000 pounds per 2,000,000 pounds of soil were added to the soils. A study was made of the effects of these applications on: (a) the number of bacteria, (b) the rate of ammonification, (c) the rate of nitrification, (d) the fixation of nitrogen by non-symbiotic organisms, and (e) the development of *B. radicicola* of the soybean. All analyses were made according to the methods given by Bear and Salter (3).

The calcium carbonate was applied and mixed thoroughly with the soil, which was then placed in 1-gallon stone jars. Enough water was added to the soil to give it an optimum moisture content. Each week the soil was removed from the jars and mixed thoroughly and the loss of moisture, due to

TABLE 2
The effect of lime on the yields of crops on soil II

PLOT	TREATMENT	YIELD PER ACRE									
		Corn 1900-1915		Oats 1901-1916		Wheat 1906-1916		Clover 1903-1916		Timothy 1909-1916	
		Unlimed	Limed	Unlimed	Limed	Unlimed	Limed	Unlimed	Limed	Unlimed	Limed
2	Phosphorus*	35.51	42.32	39.16	42.85	21.48	25.17	1848	2680	3058	3810
8	Phosphorus,* potassium.....	43.95	51.08	42.62	46.38	22.17	26.38	2144	3166	3125	3881
11	Phosphorus*, potassium, nitrogen.....	47.67	55.12	49.77	49.71	31.27	31.86	2683	3388	3445	4124
17	All three with less nitrogen but more phosphorus*....	47.23	55.67	51.84	52.38	27.32	30.85	2492	3598	3364	4543
18	Barnyard manure.....	56.31	61.68	43.62	44.93	29.51	32.49	3448	4393	4525	5531
24	Same as 17 but nitrogen in sulfate of ammonia.....	46.23	55.98	48.21	51.36	24.70	31.26	2139	3544	3111	4409
26	Same as 17 but phosphorus in bone meal.....	46.01	51.17	46.37	46.81	27.78	28.65	2945	3772	3504	4585
29	Same as 17 but phosphorus in basic slag.....	46.27	51.69	47.77	47.85	29.76	28.93	2981	3371	3741	4306
Average unfertilized.....		26.48	32.32	27.19	32.08	12.74	16.09	1276	1841	2500	3069

* Phosphorus in the form of acid phosphate.

TABLE 3
The effect of lime and acid phosphate on soil II

TREATMENT	AMOUNT PER ACRE	CORN 9 YEARS		OATS 9 YEARS		CLOVER 8 YEARS Hay
		Grain	Stover	Grain	Straw	
		pounds	pounds	pounds	pounds	
No fertilizer.....		51.50	2759	44.94	1961	4074
Calcium oxide.....	1000	57.33	3149	47.53	2079	4580
Ground limestone.....	1780	54.84	2820	45.35	1876	4362
Acid phosphate.....	320	60.18	3056	46.16	1912	4277

evaporation, was restored. This was continued for 12 weeks in order that the soil microorganisms should have time to adjust themselves to the changes in soil reaction. At the end of that time, the determinations of nitrifying power, ammonifying power, etc., were made. These determinations required about one-half of the soil.

Since the analyses showed that these soils were very deficient in total phosphorus, a thing which is commonly true of acid soils, it seemed advisable to apply phosphorus in a readily available form in order to remove it from being a possible limiting factor in the various bacterial activities studied. Accordingly, 0.2 per cent of mono-calcium phosphate, equivalent to 1000 pounds of phosphorus per 2,000,000 pounds of soil, was added, the moisture content was again restored, and the mixing was continued for another period of 12 weeks. At the end of this time, the above determinations were repeated. In some of the later experiments the calcium carbonate was added just previous to the time of studying the rate of nitrification, ammonification, etc.

THE EFFECT OF CALCIUM CARBONATE ON THE NUMBER OF BACTERIA

Soils I and II were used in these experiments, after they had received the various applications of calcium carbonate and had been mixed thoroughly each week for 12 weeks, as previously outlined. Plate counts of the number of microorganisms were made at the end of the 12-week period. After the 0.2 per cent of mono-calcium phosphate had been added and mixed with the remainder of the soil each week for a second 12 weeks, plate counts were again made. Aliquots of the soil suspension were plated on Heyden agar. The plates were incubated at room temperature and counts were made at the end of 6 days. Table 4 shows the results of these counts. Each figure represents the average of four plates.

As might be expected, the greatest relative change in the number of bacteria occurred after the neutral point had been passed. This was true in

TABLE 4
The effect of calcium carbonate on the number of bacteria

CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	BACTERIA PER GRAM OF SOIL			
	Soil I without phosphorus	Soil I with phosphorus	Soil II without phosphorus	Soil II with phosphorus
<i>pounds</i>				
0	3,341,000	4,150,000	3,503,000	3,438,000
250	4,127,000	4,320,000	3,418,000	3,536,000
500	3,537,000	3,540,000	4,614,000	4,421,000
1,000	3,439,000	2,750,000	3,781,000	5,207,000
2,000	3,930,000	2,520,000	4,472,000	5,781,000
3,000	4,127,000	4,090,000	4,919,000	5,683,000
Neutral point (Veitch method)				
4,000	4,422,000	5,820,000	7,348,000	10,005,000
5,000	5,306,000	7,700,000	9,741,000	17,392,000
7,500	5,601,000	7,070,000	15,827,000	14,297,000
10,000	3,341,000	6,680,000	14,973,000	7,959,000
20,000	6,682,000	10,750,000	14,892,000	3,635,000
40,000	9,335,000	13,050,000	18,199,000	9,826,000

both soils, as shown in figure 1. The 4000 and 5000-pound applications of calcium carbonate resulted in relatively large increases in numbers. Additions of calcium carbonate in excess of 7500 pounds per 2,000,000 pounds of soil gave somewhat uncertain results. There was a decrease in every case accompanying an application of 10,000 pounds as compared with 7500 pounds of calcium carbonate. The 20,000 and 40,000-pound applications brought about marked increases in numbers. These fluctuations were probably due to the adjustment of the soil reaction to the point where it was more suitable to the requirements of some forms which developed in vast numbers under this optimum soil reaction. It seems quite evident that the application of calcium carbonate caused decided changes in the number of bacteria in these soils. The maximum increases in numbers had apparently not been reached in three of the four cases by applications of 40,000 pounds of calcium carbonate per 2,000,000 pounds of soil. Similar trials with calcium oxide produced marked decreases in the number of bacteria in these soils following the larger applications. This was probably due to the partial sterilizing action of calcium oxide previously referred to.

THE EFFECT OF CALCIUM CARBONATE ON THE RATE OF AMMONIFICATION

Soils I and II, after the treatments with calcium carbonate and monocalcium phosphate previously referred to, were used in the experiments on ammonification. The source of nitrogen was Hammarsten's casein. Enough casein was added to supply 160 mgm. of nitrogen per 100 gm. of soil. The soil was then given an optimum moisture content and incubated in tumblers for 3 days at room temperature, after which analyses were made for ammonia by distillation with magnesium oxide. Each figure given in table 5 represents the average of two determinations which checked usually within less than 1 mgm. per 100 gm. of soil. Other determinations, not reported in this paper, were made in which only 40 mgm. of nitrogen were added, with very satisfactory results. The author believes that 160 mgm. of nitrogen per 100 gm. of soil are likely to produce abnormal conditions in a soil, although in most of the ammonification experiments reported in the literature even larger amounts of nitrogen were supplied.

The greatest relative increase in the rate of ammonification of casein, per unit of calcium carbonate applied, occurred with applications of 2000 pounds of calcium carbonate per 2,000,000 pounds of soil, as shown in figure 2. There was no marked increase in ammonification as the neutral point was passed. As previously shown, this was also the case in the number of bacteria. Applications of 250 pounds of calcium carbonate per 2,000,000 pounds of soil had a tendency to cause a decrease in the rate of ammonification. Applications of calcium carbonate in excess of 5000 pounds caused only a slight increase in the amount of ammonia produced. The 20,000 and 40,000-pound applications caused slight decreases in ammonia in several cases. There was

apparently no definite correlation between the number of bacteria and the amount of ammonia produced, although in general, increased amounts of calcium carbonate resulted in larger numbers of bacteria and more rapid ammonification.

THE EFFECT OF CALCIUM CARBONATE ON THE RATE OF NITRIFICATION

The effect of calcium carbonate on the rate of nitrification in soils I and II is shown in table 6. All figures in this and in succeeding tables of nitrification

TABLE 5
*The effect of calcium carbonate on the rate of ammonification of casein**

CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	NITROGEN AS AMMONIA PER 100 GM. OF SOIL					
	Soil I without phosphorus	Soil I with phosphorus	Soil II† without phosphorus	Soil II without phosphorus	Soil II with phosphorus	Soil II with phosphorus
pounds	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
0	72.40	60.70	38.85	22.89	29.51	39.20
250	71.00	61.00	40.25	22.05	25.55	37.38
500	72.80	59.60	45.36	29.05	28.00	37.80
1,000	75.40	62.40	48.02	29.40	28.63	44.31
2,000	78.50	68.00	57.54	41.65	43.40	55.44
3,000	79.00	70.00	56.00	44.59	45.36	57.75
Neutral point (Veitch method)						
4,000	78.40	70.60	60.41	45.85	56.84	63.00
5,000	76.30	70.80	67.27	57.12	64.61	71.54
7,500	85.30	74.80	71.40	57.05	52.29	67.97
10,000	83.60	74.80	74.48	62.79	63.07	71.61
20,000	85.40	77.80	77.40	60.76	55.27	68.81
40,000	87.50	77.60	77.42	61.25	51.61	59.36

* The results in each vertical column were obtained on the same day. Fluctuations in the temperature in the room are responsible for some of the differences observed in horizontal columns.

† Four-day periods of incubation.

represent averages of two determinations. As a rule, the duplicates agreed within less than 0.1 mgm. per 100 gm. of soil. Accordingly, only averages are reported.

These soils were treated with calcium carbonate in varying amounts and with mono-calcium phosphate as previously outlined. At the end of the 12-week periods, samples of these soils of 100 gm. each were placed in 1000-cc. Erlenmeyer flasks for the nitrification experiments. To each flask were added 20 mgm. of nitrogen in the form of either ammonium sulfate or ammonium carbonate. After adding water to the optimum content, the soils were incubated for 21 days at room temperature, after which the nitrate determinations were made by the phenol-disulphonic acid method.

A study of table 6 and figure 3 shows that the addition of calcium carbonate is followed by an increased nitrification which correlates almost directly with the increased application of calcium carbonate. This correlation holds fairly well in every case with applications up to 5000 pounds per 2,000,000 pounds of soil. There is no sudden break in the correlation as the neutral point is passed. Applications of calcium carbonate in excess of 5000 pounds are followed by increased nitrification, although the curve of increase begins to incline more toward the horizontal. In half of the experiments the curve was still ascending with applications of 40,000 pounds of calcium carbonate.

TABLE 6
The effect of calcium carbonate on the rate of nitrification

CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	NITROGEN AS NITRATE PER 100 GM. OF SOIL							
	Source of nitrogen, ammonium sulfate				Source of nitrogen, ammonium carbonate			
	Soil I* without phosphorus	Soil I with phosphorus	Soil II without phosphorus	Soil II with phosphorus	Soil I without phosphorus	Soil I with phosphorus	Soil II without phosphorus	Soil II with phosphorus
pounds	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.
0	1.08	4.06	5.28	6.07	1.38	7.22	5.29	7.50
250	1.38	4.32	4.39	5.34	1.93	8.24	5.00	8.00
500	1.43	4.60	4.40	6.38	2.11	8.42	5.42	8.40
1,000	1.82	5.24	6.15	6.75	2.21	9.52	6.60	8.55
2,000	2.29	6.38	8.50	8.73	3.01	12.42	9.03	11.55
3,000	2.96	9.34	10.48	.10.43	3.55	15.30	10.04	12.50

Neutral point (Veitch method)								
4,000	3.13	11.92	15.74	12.50	3.28	17.50	11.87	15.12
5,000	3.44	13.86	15.96	15.00	4.11	18.00	15.77	18.75
7,500	3.48	16.37	18.18	15.38	4.69	19.00	17.27	16.35
10,000	4.44	19.35	20.98	15.98	4.30	20.00	20.30	16.16
20,000	4.00	20.45	22.87	16.00	4.32	20.96	19.80	16.00
40,000	4.20	22.55	19.88	15.00	5.18	23.30	20.57	15.00

* An error was made in calculating the optimum moisture content and the soil in this experiment was too dry.

This is directly contrary to the work of Scales previously referred to, which indicated that 50 per cent of the amount of calcium carbonate necessary to supply the lime requirement of the soil is sufficient to attain the maximum rate of nitrification. Additional amounts are reported to have acted injuriously.

The marked increase in the rate of ammonification observed with the addition of 2000 pounds as compared to 1000 pounds of calcium carbonate per 2,000,000 pounds of soil was not followed by a corresponding increase in the nitrification. No correlation was found between the increased number of bacteria in the soil and the rate of nitrification except that in general the

application of increased amounts of calcium carbonate caused an upward tendency in the number of bacteria, as well as in the rate of nitrification. Since the agar plate method is not designed to include the nitrifying bacteria, no data are available as to the actual number of nitrifying organisms which were present in the soils following the applications of varying amounts of calcium carbonate.

EFFECT OF CALCIUM CARBONATE ON THE RATE OF NITROGEN FIXATION BY NON-SYMBIOTIC SOIL ORGANISMS

Samples I and II were employed again in these experiments after they had been treated as previously described. Shallow dishes having a depth of about 3 inches and a capacity of 400 gm. of soil were used for this work. Soil

TABLE 7
The effect of calcium carbonate on nitrogen fixation by non-symbiotic soil organisms

CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	NITROGEN FIXED PER 100 GM. OF SOIL		
	Soil I without phosphorus	Soil II without phosphorus	Soil II with phosphorus
pounds	mgm.	mgm.	mgm.
0	0.7	0.3	2.0
250	0.3	0.8	3.6
500	0.5	0.8	3.6
1,000	1.0	0.6	4.6
2,000	0.7	1.4	3.8
3,000	0.5	2.3	11.3

Neutral point (Veitch method)			
4,000	0.1	2.8	12.2
5,000	1.8	4.1	12.7
7,500	2.0	6.0	15.8
10,000	1.5	5.8	10.1
20,000	2.1	4.5	12.6
40,000	1.4	4.4	9.9

from the various pots to which the calcium carbonate and mono-calcium phosphate had been applied was placed in the dishes and mixed thoroughly with 2 per cent of mannit. Optimum moisture conditions were secured and maintained as nearly as possible by adding water twice daily to restore that lost by evaporation. The soils were incubated 21 days at room temperature. After thorough drying, the entire samples were pulverized to pass a 100-mesh sieve and the total nitrogen was determined in triplicate. The triplicates agreed usually within 0.05 mgm. of nitrogen on 10-gm. samples.

From a study of table 7, it appears evident that both calcium carbonate and mono-calcium phosphate were essential to the highest fixation of nitrogen. The mono-calcium phosphate had such a marked effect in increasing the nitrogen-fixing power of soil II, as shown in figure 4, that it would seem that phos-

phorus was equally as important as lime for the nitrogen-fixing organisms in this soil. The largest relative increase in nitrogen fixation followed an application of 3000 pounds of calcium carbonate per 2,000,000 of soil when accompanied by the use of mono-calcium phosphate. Heavier applications of calcium carbonate caused an increase in nitrogen fixation until as much as 10,000 pounds per 2,000,000 pounds of soil had been applied. This amount and heavier applications caused a decrease in nitrogen fixation. Apparently phosphorus was a limiting factor in nitrogen fixation in soil I, although time did not permit an experimental test of this point. The good effects resulting from the use of acid phosphate on these soils under field conditions may be due in part to this increased nitrogen fixation accompanying its use. This again is indicated by the analyses of the West Virginia Station fertility plots from which the author and others have shown that the plot receiving acid phosphate and sulfate of potash has accumulated 1173 pounds of nitrogen per acre during the last 15 years which could not be accounted for except by nitrogen fixation from the air. The evidence shown in table 7 indicates that calcium carbonate is necessary in addition to the phosphorus for the most effective nitrogen fixation.

Following the suggestion of Christensen and Larsen (4), soil from each of the pots to which varying amounts of calcium carbonate had been applied, was used to inoculate Ashby's solution in order to study the relation between the film development on the surface of the liquid and the lime requirement of the soil. The brownish film was very well developed in the flasks inoculated with soil which contained an amount of calcium carbonate in excess of the requirement of the soil and practically disappeared as the quantity of calcium carbonate applied was reduced below the amount necessary to satisfy the requirement of the soil. The development of brown pigment is apparently closely related to the amount of lime in the soil and may be used as an index of the need of lime by the soil. Other experiments which will be discussed later indicated, however, that nitrogen fixation in Ashby's solution may take place when the solution is inoculated with soils having calcium-carbonate requirements as high as 4600 pounds per 2,000,000 pounds of soil. Apparently, the lack of development of the brownish film is not accompanied by the loss of ability to fix nitrogen, since no soil was found which did not show nitrogen fixation when inoculated into Ashby's solution and allowed to stand for 21 days at room temperature.

THE EFFECT OF CALCIUM CARBONATE ON THE FIXATION OF NITROGEN BY *B. RADICICOLA* OF THE SOYBEAN (*SOJA MAX PIPER*)

Soils I and II were again used in these experiments. One-gallon pots were filled with these soils and the calcium carbonate was added. Each pot received an application of 0.2 per cent of mono-calcium phosphate. The pots were planted to soybeans, the beans having been previously inoculated with

B. radicicola. Six beans were planted in each pot and later thinned to three per pot. After the beans had reached the stage where pods were formed, they were harvested, and records were taken of their green and dry weight, the number of nodules, the dry weight of nodules and the milligrams of nitrogen in the roots, tops and nodules. One crop was harvested from each pot during the summer of 1915 and another crop during the summer of 1916. The records are shown in tables 8 and 9.

The number of nodules had a tendency to increase slightly with small applications of calcium carbonate. Applications of more than 3000 pounds of calcium carbonate per 2,000,000 of soil caused a decrease in the number of nodules. This decrease was proportional to the amount of calcium carbonate applied. The dry weight of nodules was also decreased with large applications of calcium carbonate. The rate of decrease in dry weight of nodules with increased amounts of calcium carbonate was more marked than the rate of decrease in the number of nodules. The amount of nitrogen in the nodules was almost directly correlated with the dry weight of nodules, and decreased with additional quantities of calcium carbonate. It will be noticed that in both cases the dry weight and total nitrogen of both stems and roots had a tendency to increase with small applications of calcium carbonate, but that applications in excess of 2000 pounds per 2,000,000 of soil had a tendency to cause a decrease in dry weight and total nitrogen of the stems and roots.

The total nitrogen fixed by soils I and II during the two years in which the two crops of soybeans were grown was determined. Analyses of the soil were made before and after the beans were grown. The difference in the nitrogen content of the soil at these two periods plus the nitrogen removed in the nodules, stems and roots, after subtracting the nitrogen content of the seed and water used in watering the plants, represents the nitrogen secured from the air.

The total nitrogen fixed in two years per 2,000,000 pounds of soil, as shown in the last columns of tables 8 and 9, indicate that soil II has had a more active nitrogen-fixing flora than soil I. In so far as the chemical composition is concerned, the two soils correspond fairly well, as will be found by referring to the analyses of these two soils previously shown. By referring again to table 7, showing the rate of nitrogen fixation by *Azotobacter*, it will be seen that soil II was much more active in this respect than soil I. It is possible that a greater part of the nitrogen accumulated in soil II during the growing of the legumes was fixed in the soil through the agency of the non-symbiotic organisms. The nitrogen fixation had a tendency to decrease with applications of calcium carbonate in excess of 2000 pounds per 2,000,000 pounds of soil, although the lime requirement of both soils indicated a need of 3500 pounds. Apparently, with increased applications of calcium carbonate the rate of nitrification was so high, as indicated in table 6, that the soybeans were able to secure a greater part of their nitrogen in the form of nitrates. Large numbers of *B. radicicola* were present in all the pots whether treated with

TABLE 8

*The effect of calcium carbonate on nitrogen fixation by *B. radicicola* of the soybean in soil I*

POT	CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	NUMBER	NODULES		STEMS		ROOTS		SOIL		
			Dry weight	Total nitrogen	Dry weight	Total nitrogen	Dry weight	Total nitrogen	Nitrogen in beginning per pot	Nitrogen at end per pot	Nitrogen fixed per 2,000,000 pounds of soil
	pounds		mgm.	mgm.	grams	mgm.	grams	mgm.	grams	grams	pounds
1	0	113	653	31	13.2	348	4.3	40	3.0009	2.8985	93
2	250	67	887	39	14.2	377	5.1	42	3.0039	2.8675	98
3	500	88	749	33	14.3	368	4.4	39	3.0039	2.8985	107
5	2,000	100	1017	47	17.3	464	3.7	35	3.0039	2.8272	130
6	3,000	72	560	27	14.9	303	3.4	38	3.0039	2.7900	+8

Neutral point (Veitch method)

7	4,000	65	317	16	11.9	342	2.9	37	3.0039	2.7683	-6
8	5,000	79	537	26	14.1	363	3.6	39	3.0039	2.8923	94
9	7,500	84	464	22	13.9	426	3.8	46	3.0039	2.7218	23
10	10,000	66	212	11	11.2	342	3.6	52	3.0039	2.6660	-60
11	20,000	45	220	13	10.3	298	5.1	58	3.0039	2.6815	-87
12	40,000	57	221	13	12.2	383	3.4	50	3.0039	2.6505	-56

0.1727 gm. of nitrogen in the soybeans planted.

0.0043 gm. of nitrogen in the water used in watering the soybeans.

TABLE 9

*The effect of calcium carbonate on nitrogen fixation by *B. radicicola* of the soybean in soil II*

POT	CALCIUM CARBONATE PER 2,000,000 POUNDS OF SOIL	NUMBER	NODULES		STEMS		ROOTS		SOIL		
			Dry weight	Total nitrogen	Dry weight	Total nitrogen	Dry weight	Total nitrogen	Nitrogen in beginning per pot	Nitrogen at end per pot	Nitrogen fixed per 2,000,000 pounds of soil
	pounds		mgm.	mgm.	grams	mgm.	grams	mgm.	grams	grams	pounds
1	0	79	781	38	14.8	394	4.1	46	2.6181	3.2008	570
3	500	75	744	37	15.2	440	4.0	42	2.6181	3.1860	587
6	3,000	148	577	29	13.5	371	4.7	45	2.6181	3.1418	511

Neutral point (Veitch method)

8	5,000	71	330	18	12.3	338	3.2	41	2.6181	3.1358	476
9	7,500	85	251	13	12.7	370	3.8	45	2.6181	3.1270	490
10	10,000	66	207	12	11.8	335	3.3	57	2.6181	3.0238	408
11	20,000	54	124	5	11.5	341	4.0	68	2.6181	2.9648	377
12	40,000	33	126	7	14.4	393	3.0	40	2.6181	2.9205	265

0.1727 gm. of nitrogen in the soybeans planted.

0.0043 gm. of nitrogen in the water used in watering the soybeans.

calcium carbonate or not, although quantitative determinations were not made of their numbers.

THE EFFECT OF CALCIUM CARBONATE ON SOYBEANS UNDER FIELD CONDITIONS

In order to determine whether soybean yields are increased by the use of calcium carbonate on soil I under field conditions, it was decided to grow soybeans on the fertility plots of the station farm during the summer of 1916. Three varieties of soybeans were sown in rows across the plots and cultivated during the growing season. One-half of each plot received an application of calcium carbonate at the rate of 2 tons per acre in the form of ground limestone. The yields of hay produced are given in table 10. The previous crop records and the analyses of the soils of these plots are given in tables 1 and 11.

TABLE 10
Effect of ground limestone on yield of soybean hay on soil I

PLOTS	TREATMENT	CALCIUM-CARBONATE REQUIREMENT PER 2,000,000 POUNDS	YIELD OF HAY PER ACRE		INCREASE WITH LIMESTONE
			No limestone	Limestone	
19	N, P, K, CaO.....	0	5270	5400	+2
20	M, CaO.....	0	6390	6850	+7
21	Check.....	2800	1605	1400	-13
22	CaO.....	0	1920	2430	+26
26	M.....	2800	7150	7360	+3
26	N, P, K.....	3200	5300	5370	+1
28	P, K.....	3600	2820	4690	+66
29	N, K.....	3400	1285	2280	+77
31	N, P.....	3200	3375	4460	+32
32	K.....	3600	1495	1995	+34
34	P.....	3400	3285	4105	+25
35	N.....	3400	1220	1705	+40
Averages.....			3426	4004	+17

From a study of the plots and the crop records, it would seem that the use of 2 tons of limestone per acre did not give sufficient increase in yield to justify the conclusion that soybeans will not grow well except on soils which have had their lime requirement satisfied. On plots 25 and 26, the soils of both of which have rather high calcium-carbonate requirements, but which also contain a fairly high content of nitrogen, the yield of soybeans was little affected by the limestone. This might mean that more nitrogen was secured from the soil on these plots and for this reason the crop was larger. However, the nodules were plentiful on the roots of the soybeans on plots 25 and 26 and, therefore, we could assume that nitrogen fixation from the air was taking place.

EFFECT OF CALCIUM CARBONATE ON THE BACTERIAL ACTIVITIES OF DEKALB SOILS HAVING VARYING LIME REQUIREMENTS

A large number of samples of acid soils all belonging to the Dekalb series were chosen from various parts of West Virginia and sent to the laboratory. From this number 12 samples were chosen which had calcium-carbonate requirements varying from 400 to 4600 pounds per 2,000,000 pounds of soil. The analyses of these soils are shown in table 11. These soils differ mostly because of the different systems of management practiced by the men who have farmed them since the areas from which the samples were chosen were cleared from the forest. Many of these areas had been farmed for from seventy-five to one-hundred years and others had not been farmed for more than a few years.

TABLE 11
Analyses of soils of table 12

SAMPLE	POUNDS PER 2,000,000 POUNDS OF SOIL			
	Nitrogen	Phosphorus	Carbon	Calcium-carbonate requirement
III	3870	1203	41,420	400
IV	1904	586	21,790	1000
V	1669	680	17,600	1200
VI	3374	697	47,230	1400
VII	3142	902	32,140	1600
VIII	2042	1216	20,280	2000
IX	2602	662	32,450	2200
X	4142	1135	48,680	2600
XI	3384	660	39,490	2800
XII	2750	706	32,140	3200
XIII	1960	608	21,900	3800
XIV	3124	753	48,280	4600

The rates of nitrification, ammonification, and nitrogen fixation were studied in an attempt to determine whether there was any relation between the activities of the soil organisms and the calcium-carbonate requirements of these soils.

In nitrification studies 100 gm. of soil to which varying amounts of calcium carbonate had been added were placed in 1000-cc. Erlenmeyer flasks and incubated with optimum moisture content at room temperature for 21 days, using ammonium sulfate as the source of nitrogen, adding a sufficient amount to supply 20 mgm. of nitrogen per 100 gm. of soil. In ammonification studies 100-gm. samples of soil were used and varying amounts of calcium carbonate were added as in the nitrification tests. Casein was used as the source of nitrogen, 160 mgm. of nitrogen being added to 100 gm. of soil. The soil was incubated in tumblers at optimum moisture content for 3 days and the am-

monia determined by distillation with magnesium oxide. Nitrogen-fixation tests were carried on by placing 10 gm. of soil in 100 cc. of Ashby's solution in 800-cc. Kjeldahl flasks for a period of 21 days at room temperature. To one set of flasks enough calcium-carbonate was added to be equivalent to 10,000 pounds per 2,000,000 pounds of soil. At the end of 21 days the total nitrogen was determined. All of the determinations on nitrification, ammonification, and nitrogen fixation were performed in duplicate and these duplicates as a rule checked very closely. The results of these experiments are tabulated in table 12.

In general the highest rates of ammonification occurred with soils having the lowest calcium-carbonate requirements. The applications of 2000 pounds and 5000 pounds of calcium-carbonate brought about marked increased in the rate of ammonification. Applications of 10,000 pounds of calcium carbonate per 2,000,000 pounds of soil caused a decreased ammonification except in soils XIII and XIV, which had calcium-carbonate requirements of 3800 and 4600 pounds, respectively. Apparently the application of 10,000 pounds of calcium carbonate per 2,000,000 of soil on soils having calcium-carbonate requirements of less than 3800 pounds is injurious to ammonifying organisms.

There was no very definite correlation between the rate of nitrification of ammonium sulfate and the calcium-carbonate requirement of the soils. In general, the soils having high calcium-carbonate requirements had a very low nitrifying power. With soils having calcium-carbonate requirements in excess of 2200 pounds per 2,000,000 pounds of soil, the nitrifying organisms did not become markedly active even with large applications of calcium carbonate. Either the nitrifying organisms were almost entirely absent or had become very inactive because of the unfavorableness of the medium in which they were living.

There was no very marked correlation between the calcium-carbonate requirement of these soils and the nitrogen-fixing power of the soil organisms in Ashby's solution. Soil XIV, having a calcium-carbonate requirement of 4600 pounds, was able to fix nitrogen to the extent of 2.9 mgm. per 100 cc. of Ashby's solution in 21 days. The rate of nitrogen fixation was increased in every case by the addition of calcium carbonate, but the effect was more marked on soils having a high requirement than in soils having a low calcium-carbonate requirement. It seems remarkable, however, that nitrogen fixation took place in all cases even though some of the soils had very high lime requirements.

EFFECT OF FERTILIZERS ON THE BACTERIAL ACTIVITIES OF SOILS

These experiments were conducted in order to determine what effect differences in the fertilizer treatments of the same soil would have on the bacterial activities in the soil. Samples of soil were chosen from 12 plots of the fertilizer series of the fertility plots on the West Virginia station, some of which differ considerably because of the fertilizer applications they have received during the last 15 years. Records of the treatments of the soil on these plots have

TABLE 12

The effect of calcium carbonate on the activities of soil bacteria in Dekalb soils having varying lime requirements

SOIL	CALCIUM-CARBONATE REQUIREMENT PER 2,000,000 POUNDS OF SOIL	CALCIUM CARBONATE APPLIED PER 2,000,000 POUNDS OF SOIL	NITROGEN PER 100 GM. OF SOIL				NITROGEN FIXED IN 100 CC. OF ASHBY'S SOLUTION	
			Nitrogen as ammonia from casein		Nitrogen as nitrates from ammonium sulfate			
			mgm.	relative	mgm.	relative	mgm.	relative
III	400	{ 0 2,000 5,000 10,000	68.9	100	8.0	100	7.6	100
			76.5	111	12.6	158		
			87.6	127	12.5	156		
			82.4	118	9.4	118	8.7	114
IV	1,000	{ 0 2,000 5,000 10,000	67.1	100	4.4	100	4.2	100
			81.2	121	8.4	191		
			86.4	129	13.5	307		
			74.6	111	14.0	318	5.6	133
V	1,200	{ 0 2,000 5,000 10,000	68.9	100	1.3	100	2.7	100
			80.5	117	7.8	600		
			78.1	114	12.0	969		
			72.6	105	16.0	1231	4.1	152
VI	1,400	{ 0 2,000 5,000 10,000	73.5	100	5.8	100	4.4	100
			83.2	113	7.3	126		
			86.9	118	8.0	138		
			87.7	119	12.6	217	5.6	127
VII	1,600	{ 0 2,000 5,000 10,000	56.8	100	1.5	100	4.9	100
			77.3	136	3.0	200		
			91.8	161	5.7	380		
			87.5	154	16.0	1067	5.9	120
VIII	1,800	{ 0 2,000 5,000 10,000	67.0	100	0.4	100	5.1	100
			76.2	114	1.5	375		
			96.6	129	8.0	2000		
			87.3	130	15.7	3925	6.5	127
IX	2,200	{ 0 2,000 5,000 10,000	69.6	100	0.8	100	3.1	100
			83.1	119	5.1	638		
			90.1	129	9.8	1225		
			82.7	119	4.6	575	4.7	151
X	2,600	{ 0 2,000 5,000 10,000	62.9	100	2.7	100	3.4	100
			75.1	119	4.8	144		
			92.2	147	5.2	156		
			91.8	146	13.0	390	4.8	141
XI	2,800	{ 0 2,000 5,000 10,000	51.6	100	0.8	100	2.3	100
			68.3	132	2.2	275		
			88.9	172	1.7	213		
			92.2	159	2.1	263	3.2	139

TABLE 12—(Continued)

SOIL	CALCIUM-CARBONATE REQUIREMENT PER 2,000,000 POUNDS OF SOIL	CALCIUM CARBONATE APPLIED PER 2,000,000 POUNDS OF SOIL	NITROGEN PER 100 GM. OF SOIL				NITROGEN FIXED IN 100 CC. OF ASHBY'S SOLUTION	
			Nitrogen as ammonia from casein		Nitrogen as nitrates from ammonium sulfate			
	pounds	pounds	mgs.	relative	mgs.	relative	mgs.	relative
XII	3,200	{ 0 2,000 5,000 10,000	49.6	100	0.4	100	4.4	100
			70.5	142	1.4	350		
			93.0	187	2.0	500		
			89.5	180	3.2	800	5.6	127
XIII	3,800	{ 0 2,000 5,000 10,000	46.0	100	1.1	100	5.4	100
			62.1	135	3.0	273		
			76.3	166	4.5	410		
			81.3	177	3.8	345	7.1	131
XIV	4,600	{ 0 2,000 5,000 10,000	30.0	100	0.3	100	2.9	100
			45.5	152	0.7	233		
			72.4	241	1.1	367		
			86.3	288	0.7	233	4.4	152

been given in table 1 previously referred to. Analyses of the soil on the various plots were made and are recorded in table 13.

The studies in nitrification, ammonification, and nitrogen fixation were conducted in the same manner as previously mentioned in the discussion of the 12 soils of the Dekalb series with varying calcium-carbonate requirements. It will be remembered that the soil of the fertility plots is also Dekalb soil. The records of these experiments are shown in table 14.

TABLE 13
Analyses of soils of table 14

PLOT	TREATMENT	POUNDS PER 2,000,000 POUNDS OF SOIL			
		Nitrogen	Phosphorus	Carbon	Calcium-carbonate requirement
		pounds	pounds	pounds	pounds
19	N, P, K, CaO.....	2130	765	24,500	0
20	M, CaO.....	2700	1045	32,500	0
21	Check.....	1830	590	21,200	2800
22	CaO.....	1750	510	19,400	0
25	M.....	3240	1220	36,800	2800
26	N, P, K.....	2665	900	30,400	3200
28	P, K.....	2280	850	26,000	3600
29	N, K.....	2290	640	27,000	3400
31	N, P.....	2395	880	28,000	3200
32	K.....	2310	740	29,200	3600
34	P.....	2300	885	28,200	3400
35	N.....	2100	620	28,800	3400

N indicates nitrate of soda; P, acid phosphate; K, sulfate of potash; M, manure.

TABLE 14

The effect of calcium carbonate on the activities of soil bacteria in Dekalb soils which have received varying fertilizer treatments

TREATMENT	CALCIUM-CARBONATE REQUIREMENT PER 2,000,000 POUNDS OF SOIL pounds	CALCIUM CARBONATE APPLIED PER 2,000,000 POUNDS OF SOIL pounds	NITROGEN PER 100 GM. OF SOIL				NITROGEN FIXED IN 100 CC. OF ASHBY'S SOLUTION			
			Nitrogen as ammonia from casein		Nitrogen as nitrates from ammonium sulfate					
			mgm.	relative	mgm.	relative				
N, P, K, CaO.....	0	0	78.3	100	15.3	100	5.6	100		
		2,000	80.1	102	18.5	121				
		5,000	78.3	100	19.5	127				
		10,000	77.4	99	20.3	133	7.5	134		
N, CaO.....	0	0	87.8	100	17.5	100	6.2	100		
		2,000	89.5	102	21.5	123				
		5,000	87.0	99	22.5	129				
		10,000	84.3	96	22.0	126	6.4	103		
Check.....	2,800	0	60.6	100	1.2	100	3.7	100		
		2,000	65.8	108	5.4	450				
		5,000	78.2	129	11.8	983				
		10,000	79.7	131	15.5	1275	4.2	113		
CaO.....	0	0	71.8	100	8.5	100	2.6	100		
		2,000	75.2	105	15.8	187				
		5,000	82.9	115	18.3	215				
		10,000	82.7	115	22.0	259	4.8	185		
M.....	2,800	0	71.7	100	6.7	100	5.1	100		
		2,000	75.3	105	12.5	186				
		5,000	90.5	126	16.3	243				
		10,000	90.0	125	21.5	321	7.2	141		
N, P, K.....	3,200	0	70.1	100	2.9	100	4.2	100		
		2,000	80.3	114	7.0	241				
		5,000	85.5	122	9.3	321				
		10,000	86.6	126	13.8	475	6.1	145		
P, K.....	3,600	0	58.4	100	1.4	100	4.3	100		
		2,000	66.2	109	4.3	301				
		5,000	76.3	131	8.5	601				
		10,000	82.4	141	13.0	928	5.5	128		
N, K.....	3,400	0	56.6	100	1.5	100	5.1	100		
		2,000	65.9	116	4.8	320				
		5,000	75.5	133	7.0	467				
		10,000	81.8	144	10.0	667	6.8	133		
N, P.....	3,200	0	60.9	100	1.8	100	4.4	100		
		2,000	68.7	113	5.8	322				
		5,000	82.9	136	9.7	504				
		10,000	85.2	140	12.8	701	5.5	125		

TABLE 14—(Continued)

TREATMENT	CALCIUM-CARBONATE REQUIREMENT PER 2,000,000 POUNDS OF SOIL	CALCIUM CARBONATE APPLIED PER 2,000,000 POUNDS OF SOIL	NITROGEN PER 100 GM. OF SOIL				NITROGEN FIXED IN 100 CC. OF ASHBY'S SOLUTION	
			Nitrogen as ammonium from casein		Nitrogen as nitrates from ammonium sulfate			
	pounds	pounds	mgm.	relative	mgm.	relative	mgm.	relative
K.....	3,600	0	47.9	100	1.1	100	7.4	100
		2,000	57.4	120	2.5	237		
		5,000	75.7	158	5.0	454		
		10,000	84.0	174	6.6	600	7.6	103
P.....	3,400	0	49.5	100	1.1	100	5.3	100
		2,000	65.0	131	3.4	301		
		5,000	79.4	160	7.0	636		
		10,000	84.3	170	8.5	772	7.6	143
N.....	3,400	0	50.2	100	1.1	100	3.8	100
		2,000	64.9	129	2.9	272		
		5,000	78.6	156	5.5	500		
		10,000	83.1	165	7.3	663	6.9	182

Nitrification of ammonium sulfate was not very active in these soils except on the plots where lime had been applied in the field. Even the soil of plots 26, 28 and 31, which had been producing very satisfactory crops as indicated in table 1, did not contain vigorous nitrifying organisms. The rate of nitrification was materially increased by applications of calcium carbonate. The nitrifying organisms were much more active in the soil from the manure plots than in the soil from any of the other plots except where lime had been applied. There was a general tendency for the rate of ammonification of casein to decrease with an increase in the lime requirement of the soils. There were some marked exceptions to this tendency, notably plots 25 and 26. A study of the analyses of these plots shows a high total content of nitrogen and organic matter. No lime has ever been applied to plots 25 and 26. This increased nitrogen in the form of protein represents an increased amount of material available for the action of ammonifying organisms. If a large amount of nitrogen has been stored up in the soil, the amount of ammonia produced without any applications of calcium carbonate would be sufficient to produce satisfactory yields of those crops which are able to utilize ammonia, on a soil having lime requirements no higher than those of plots 25 and 26. The tendency for small applications of calcium carbonate to be relatively much more effective than larger applications was again shown in these experiments. It was evident that ammonification proceeds fairly satisfactorily without the application of calcium carbonate, especially, as suggested in the preceding discussion, if the content of organic nitrogen is high. Large applications of calcium carbonate had a tendency to reduce the rate of ammonification.

Nitrogen fixation took place very readily in Ashby's solution when inoculated with soil from any of the plots. There did not seem to be any correlation between the calcium-carbonate requirement and nitrogen fixation. The addition of calcium carbonate to Ashby's solution caused an increase in nitrogen fixation in every case, but this increase was no more marked in soil having a high lime requirement than in soil having a low lime requirement.

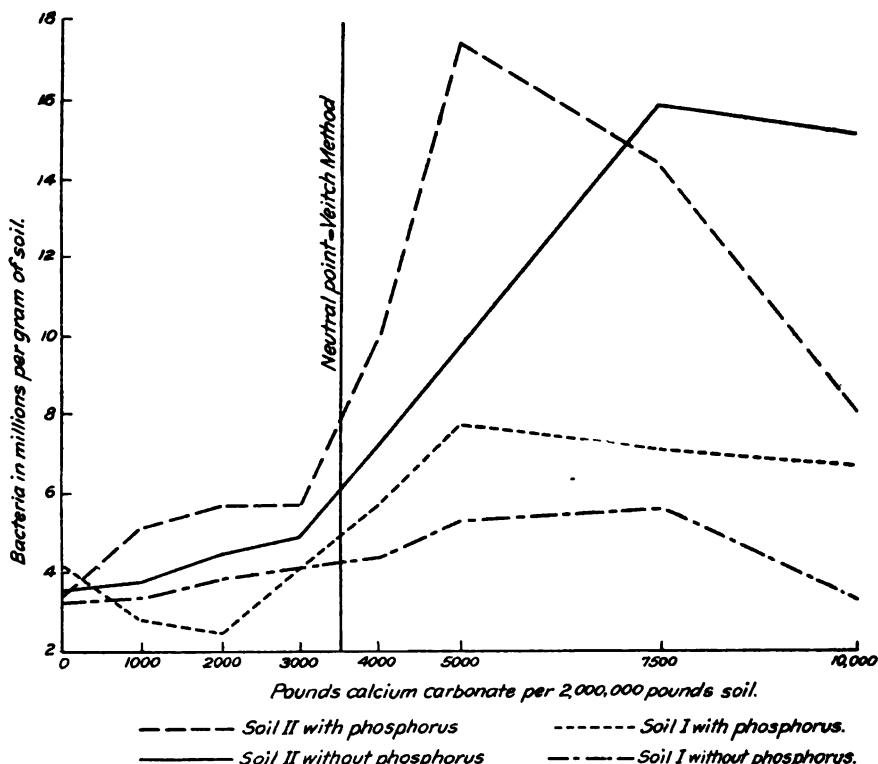


FIG. 1. THE EFFECT OF CALCIUM CARBONATE ON THE NUMBER OF BACTERIA IN SOILS I AND II

SUMMARY AND CONCLUSIONS

This investigation was undertaken as a preliminary step in the study of the possibilities of a system of acid agriculture on soils somewhat distantly removed from a source of lime. A study was made of the relation between the activities of the soil bacteria concerned in nitrogen accumulation and nitrogen transformations and the lime requirement of certain soils. The lime requirement of these soils varied from none to 4600 pounds of calcium carbonate per 2,000,000 pounds of soil. To different portions of these soils calcium carbonate was added in amounts ranging from 0.01 per cent to 2 per cent of the weight

of the soil. The data accumulated show that the various groups of soil organisms vary in their response to applications of calcium carbonate.

Ammonification proceeded fairly satisfactorily in most of the soils without the application of lime. The use of moderate amounts of calcium carbonate increased the rate of ammonification in most cases. Small applications were much more effective, relatively, than large applications.

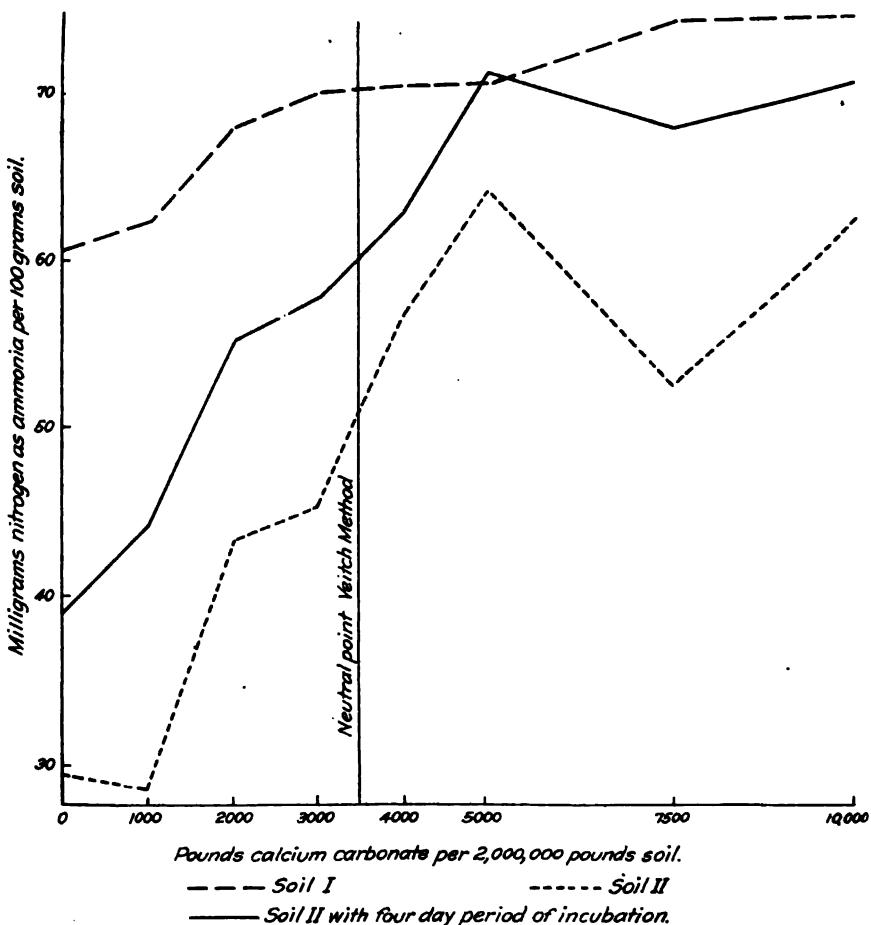


FIG. 2. THE EFFECT OF CALCIUM CARBONATE ON THE RATE OF AMMONIFICATION IN SOILS I AND II WITH PHOSPHORUS

The rate of nitrification was almost directly correlated with the amount of calcium carbonate supplied. Excessive applications were not injurious to the nitrifying organisms. Soils having high lime requirements showed practically no nitrification until calcium carbonate had been mixed with them.

Nitrogen fixation by non-symbiotic soil organisms was considerably in-

creased by the addition of calcium carbonate. The application of monocalcium phosphate also was necessary for maximum nitrogen fixation. All of the soils studied accumulated considerable amounts of nitrogen when incubated in Ashby's solution without the addition of calcium carbonate, although its use increased the rate of nitrogen fixation.

A lime requirement of 3000 pounds was not sufficient to prevent a good growth of soybeans on soil well fertilized with acid phosphate or manure. Nitrogen fixation accompanying the growth of soybeans took place readily

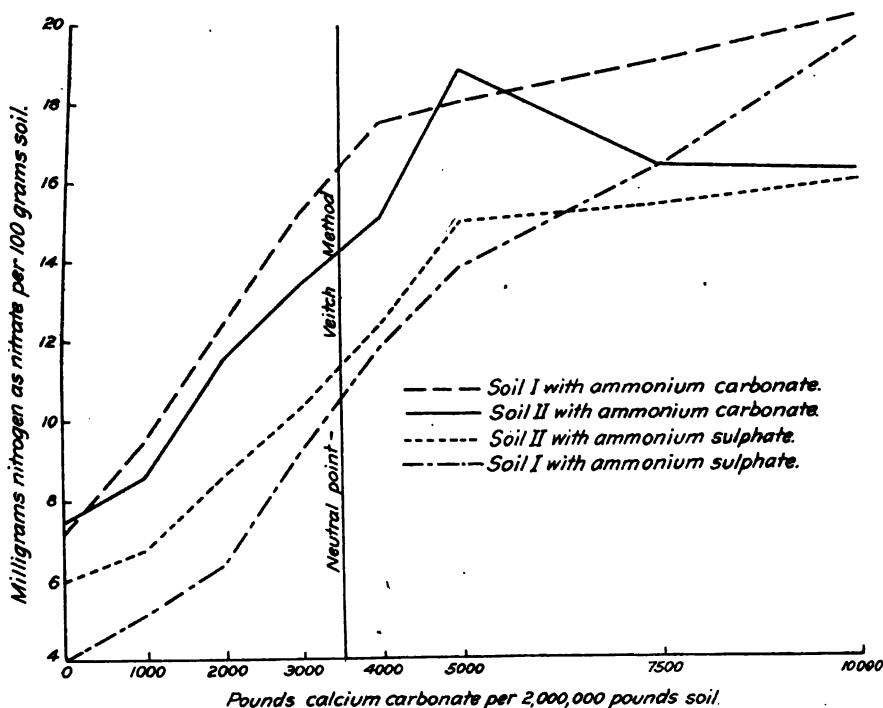


FIG. 3. THE EFFECT OF CALCIUM CARBONATE ON THE RATE OF NITRIFICATION IN SOILS I AND II WITH PHOSPHORUS

in acid soils. This fixation was increased by small applications but decreased by large applications of calcium carbonate.

From these facts the following conclusions seem justified:

1. Plants which are able to utilize ammonia nitrogen need not suffer from nitrogen hunger when grown on soils having lime requirements no higher than those studied in these investigations.
2. Plants which depend on nitrates as their source of nitrogen may suffer from the lack of available nitrogen in soils having high lime requirements, unless these requirements have been at least partially satisfied.

3. The supply of nitrogen in acid soils may be maintained by growing acid-resistant legumes, of which the soybean is one. Undoubtedly, the use of acid phosphate aids materially in the nitrogen-fixation processes in acid soils.

4. Small applications of calcium carbonate are, as a rule, relatively more effective than large applications as a means of increasing the bacterial activities in acid soils.

Acknowledgment is due Dr. E. B. Fred of the University of Wisconsin for many helpful suggestions and criticisms offered during the progress of this investigation.

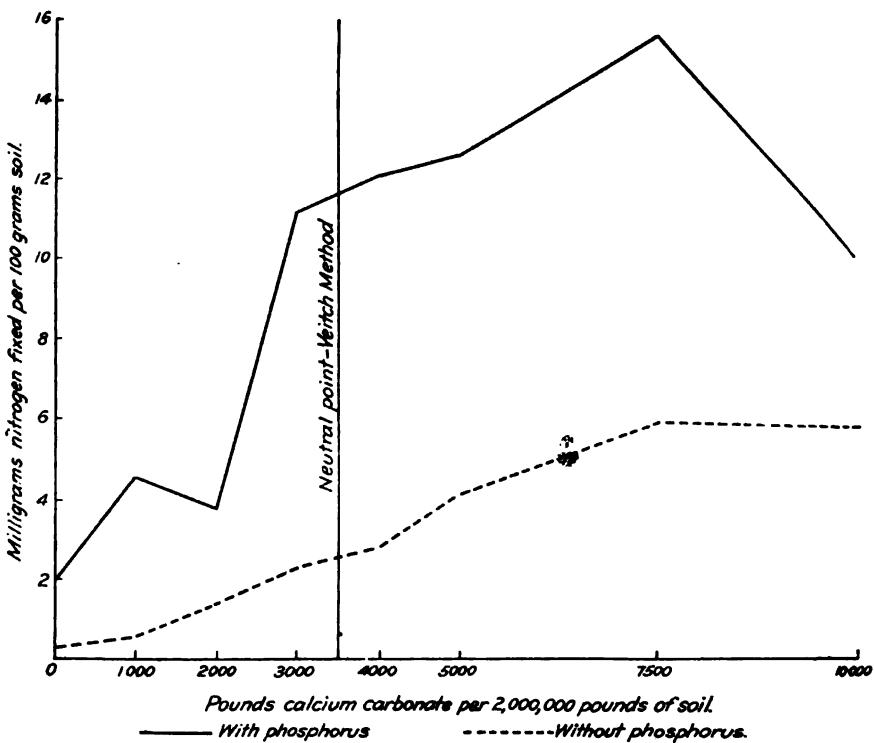


FIG. 4. THE EFFECT OF CALCIUM CARBONATE ON NON-SYMBIOTIC NITROGEN FIXATION IN SOIL II

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THE MOISTURE EQUIVALENT DETERMINATIONS OF SALT-TREATED SOILS AND THEIR RELATION TO CHANGES IN THE INTERIOR SURFACES

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The physical properties of soils in the field or laboratory may be appreciably modified by the addition of certain acids, bases, and salts. It is also true that a still more pronounced change in the soil properties will result if some of these added substances are washed either wholly or partly from the soil with water. To be consistent with the more recent theoretical reasoning, one must regard the changes in the degree of dispersion of the soil particles and the consequent alteration in the interfacial surfaces as important effects of such salt treatments. The more commonly noted effects as the changes in permeability, capillary activity, response to cultivation, and other similar soil factors are, we believe, to be properly regarded as incidental to and dependent upon the effects mentioned above.¹ Frequently, the exact relationship between these more obvious effects of salt treatments and the changes brought about in the interior soil surface are obscured by factors, the expressions for which have not been established. It is evident that those methods which most closely approach, either directly or indirectly, the measurement of the interior surface of soils are the most suitable for studies concerned with this phase of soil physics. Among the several methods which have been used in this laboratory for this purpose, the centrifugal determination of the moisture equivalent described by Briggs and McLane (1) seems to be one of the most promising.

Briggs and Shantz (2) have already pointed out the existence of important relationships between the moisture equivalent and the hygroscopic coefficient, the water-retaining power, and the mechanical analysis of soils. In addition they have correlated these coefficients with the wilting coefficient. Thus a single determination provides a comparatively simple, convenient, and fairly accurate means of connecting the physical properties of soils with their effects on biological processes. These reasons are sufficient to warrant a study of the salt effects on soils as indicated by the moisture-equivalent determinations. The first part of this paper presents the data secured by us

¹ Any effects which might be directly attributed to changes in the physical properties of the liquid phase, i.e., surface tension, vapor pressure, and density, are not considered in this discussion. We are dealing here with the changes in the solid phase, the soil particles.

in our studies. Aside from its usefulness in the directions already referred to, the moisture equivalent may be a means of obtaining a clearer conception of the comparative magnitudes of the interior surfaces of soils. At present this is purely a theoretical consideration and the few words devoted to it in the second part of this paper will suffice to explain it.

EXPERIMENTAL DATA

Duplicate 100-gm. portions of the Davis clay loam soil, which had passed the 2-mm. sieve were treated with 80 cc. of the salt solutions of the concentrations specified in table 1. From one of these portions the salt was washed with distilled water before centrifuging, while a sample from the other portion was subjected to centrifuging without washing. In numbers 19 to 21, inclusive, a sufficient quantity of the various salts was added to yield an amount of base equivalent to 3 gm. of sodium to 100 gm. of soil. Table 1 gives an outline of these treatments and the corresponding moisture equivalents.

The data given in table 1 show that the addition of NaCl and Na₂SO₄ to the Davis soil did not materially modify the moisture equivalent. In all probability the explanation of this lies in the fact that the smaller soil particles are flocculated to such a degree under normal conditions that the further addition of the flocculants NaCl and Na₂SO₄ proved to be without measurable effect on the moisture equivalent. On the other hand, it is likely that if a soil, the particles of which were in a state of diffusion, were substituted for the Davis soil, then more noticeable effects would result from the application of flocculating agents.

To formulate an explanation for the behavior of the Davis soil to which Na₂CO₃ and NaOH had been added is not as simple as in the case of the neutral salts. We would expect that these alkaline substances would increase, very markedly, the degree of dispersion of the soil particles. That this is not always the case is evident from some other experiments in which Na₂CO₃ (3) under certain circumstances was shown to lack the deflocculating power exhibited by NaOH toward suspensions of the Davis soil. This probably accounts for the similarity of the moisture equivalent of the soil treated with Na₂CO₃ and that of the control soil in the present experiments. As previously mentioned, NaOH of certain concentrations increases the degree of dispersion of the soil particles. A corresponding increase in the moisture equivalents of the soils treated with the proper concentrations of NaOH should logically follow. On the contrary, our experiments seem to indicate that the moisture equivalent is not changed by the addition of NaOH. We are not, at this time, in a position to offer a satisfactory explanation of this behavior. On the whole, it can be said that while the added salts are present in the soil, little or no change in the moisture equivalent was observed.

A very different effect is produced if these same salts are washed from the soil with water. The soils so treated seem to possess a new and peculiar set

TABLE I

Effect of salts and washing treatments on the moisture equivalent of the Davis soil

NUMBER	SALT	CONCENTRATION	SALT ADDED PER 100 GM. OF SOIL	MOISTURE EQUIVALENTS			
				' Salt present		Salt leached out	
				Duplicates	Average	Duplicates	Average
grams							
1	00	00	00	19.06	19.26	19.34	19.15
3	NaCl	N	4.680	20.10	19.70	24.26	24.94
4	NaCl	N/10	0.468	19.20	19.60	22.08	22.14
5	NaCl	N/50	0.093	19.00	19.45	22.29	22.35
6	NaCl	N/100	0.046	20.10	19.70	21.53	21.46
7	Na ₂ SO ₄	N	5.680	19.00	19.10	32.19	32.05
8	Na ₂ SO ₄	N/10	0.568	19.80	19.75	22.97	22.89
9	Na ₂ SO ₄	N/50	0.113	19.70	19.85	21.45	21.53
10	Na ₂ SO ₄	N/100	0.056	20.20	20.15	20.73	20.57
11	Na ₂ CO ₃	N	4.240	19.62	19.63	29.45	28.90
12	Na ₂ CO ₃	N/10	0.424	20.01	20.02	20.03	20.01
13	Na ₂ CO ₃	N/50	0.085	18.40	18.53	19.05	19.05
14	Na ₂ CO ₃	N/100	0.042	18.86	18.80	18.69	18.69
15	NaOH	N	3.200	19.92	20.01	29.70	30.01
16	NaOH	N/10	0.320	20.52	20.29	22.24	24.18

TABLE 1 (Cont.)

No.	SALT	CONCENTRATION	SALT ADDED PER 100 GM. OF SOIL	MOISTURE EQUIVALENTS			
				Salt present		Salt leached out	
				Duplicates	Average	Duplicates	Average
17	NaOH	N/50	0.064	19.24 19.37	19.30	19.44 19.24	19.34
18	NaOH	N/100	0.032	18.58 18.73	18.65	19.43 19.44	19.43
19	NaNO ₃	Salt added equivalent to 3 gm. of base per 100 gm. of soil	11.078			30.96 30.32	30.64
20	NaC ₂ H ₅ O ₂		15.620			32.86 29.60	31.23
21	CaCl ₂		8.300			19.88 19.22	19.55

of physical properties. As shown in table 1 the moisture equivalent of the Davis soil is markedly increased by such treatments. The extent to which the moisture equivalent is changed depends upon the salt used. Thus the washing out of all the sodium salts was accompanied by a considerable increase in the moisture equivalent, while the washing out of CaCl₂ did not perceptibly alter this factor. Since the leaching out of other salts as KCl, K₂SO₄, KNO₃ and (NH₄)₂ SO₄ produces an effect quite similar to that existing after the sodium salts have been leached from the soil, it is highly probable that the washing out of the salts first mentioned would produce effects on the moisture equivalent commensurate with those found when sodium salts have been leached from the soil.

The amount of change in the moisture equivalent due to leaching seems to depend also upon the anion with which the sodium is associated. In the experiment reported Na₂SO₄ produced the greatest effect, followed in order by NaOH, Na₂CO₃ and NaCl. Of course, equal quantities of sodium are under comparison.

The absolute quantity of salt with which the soil has been treated is likewise an important factor in determining the extent to which the soil will be modified. The larger applications followed with washing of salts invariably produced the greater effects on the moisture equivalent. The two smaller applications of Na₂CO₃ and NaOH were without a measurable effect. The application of some of the results of the investigations of Briggs and Shantz to our own problem of handling the salt-treated, water-washed soils has enabled us to see more clearly the reasons for the difficulties involved in

attempting to grow plants in such diffused soils, and have offered us valuable suggestions as to the proper procedure for alleviating these conditions.

THEORETICAL DISCUSSION

The optimum physical conditions for plant growth in such heterogeneous mediums as the soil mass must obviously depend in some measure upon the interfaces between the various phases and the factors affecting them. As mentioned in the first part of this paper, one of these factors is the effect of salts on the degree of dispersion of the soil particles. Furthermore, in many cases the leaching out of salts is instrumental in bringing about a greater change in the degree of dispersion of the soil particles than that produced by the mere addition of the salts, and hence it would seem that the salt and water treatments would also be accompanied by greater changes in the interfacial surfaces. These statements serve to introduce some of the intricacies of the problems involved in the measurement of surfaces within the soil mass, especially those affected by salts.

We now propose to consider the moisture equivalent determinations as functions of the interior surfaces of soils. Our assumption presupposes that the moisture equivalent varies directly as the surface of the solid soil particles. Such a relationship would be the logical deduction from the following considerations.

From the moisture equivalent it is possible to obtain the volume of water which a given quantity of soil will retain against a given force. According to Briggs and McLane (1) the water so held is in capillary equilibrium. That is to say, the water films in any two soils, which have been centrifuged, are of equal thickness and the liquid-air surfaces in the capillary spaces are of the same curvature.

By momentarily neglecting the possibility that the only inequality that any two soils may show with respect to their residual water content after centrifuging, lies in the number of points of contact between particles, or the number of capillary spaces, we are enabled to consider the water usually designated by the term "moisture equivalent" to be spread over the surfaces of the two soils to an equal depth, and hence the surfaces would vary directly as the volume of water retained.

Thus let X be the thickness of the water film and A and B the volumes of water retained by two soils. Then the surfaces S_a and S_b are as follows: $S_a = A/X$ and $S_b = B/X$, and S_a is to S_b as A is to B . If a finite value could be assigned to X , then the figures for S_a and S_b would approach absolute values.

The most serious objection to such a line of reasoning lies in the factor which we purposely neglected for the moment, namely, the number of capillary spaces in which water may collect to a depth greater than that in the surface films. Obviously, in such cases the thickness of the water layers

would not be uniform throughout, which possibly invalidates any rigorous mathematical interpretation of the data obtained by the use of the centrifuge. On the other hand, the differences in thickness may not be so great as to militate seriously against the correctness of the general proposition that the moisture equivalent is a more or less accurate function of the interior surface.

But there are certain compensating factors which would tend to equalize the thickness of the water layers, even though the number of points of contact between particles should be increased. This can best be shown by the following sketch:

In *A* two spherical particles are shown suspended in a liquid some of which has become closely associated with the surfaces of the particles. If the excess liquid is gradually removed the particles come closer together and the liquid films around them assume a form as in *b*. In the immediate region between the two spheres the thickness of the water films is somewhat decreased, while further out from the spheres the film thickness is increased. *C* represents the final appearance of such a system when the excess water is removed by some force as that developed by centrifuging. In this case the

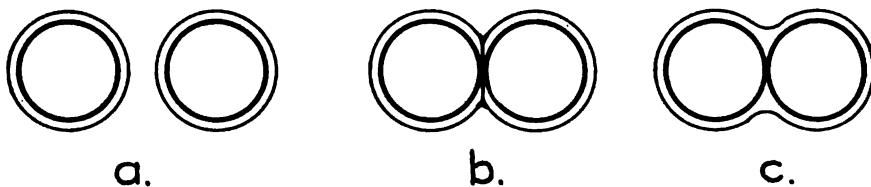


FIG. 1

particles are in contact with each other, necessitating a marked decrease in the thickness of the film adjacent to the point of contact. A further examination of this system shows, however, that this decrease in thickness may be largely compensated for by a corresponding increase in the thickness of the water layers in the capillary spaces. In other words, the total volume of water retained and the actual surface of the particles are not materially changed by increasing or decreasing the number of points of contact between the particles. Hence, the average thickness of the water film surrounding the soil particles would likewise be undisturbed. Evidently, the extent of the surface over which the residual water is to be spread is of more importance in determining the volume of water retained than the number of capillary spaces. The latter statement refers to soils having particles of the same size but differing in the arrangement of these particles. Of course, any change in the actual size of the particles or even in their effective size necessarily means change in the interior surface of the soil, and hence a change in the moisture equivalent. It is for the reasons outlined above that the moisture equivalent is considered as an index of the interior surface of soils.

A second objection to our proposal lies in the fact that the conditions of an ideal system composed of spheres of the same size and pure water are not fulfilled by the soil system, for the soil particles are not spherical, nor are they of uniform size; hence the laws concerning the first type of system may not hold with the same exactitude for the soil-water-air system. Although the soil particles may be variously shaped, the indentures and other irregularities on their surfaces would act just as points of contact accompanied with interstitial spaces, so that, in the main, the surface of the particle would be the primary factor in the retention of water against any force.

SUMMARY

With these points in view we may conclude that the salt and water treatments have increased the interior surface of the soils from 2 to 40 per cent, the magnitude of the increase depending upon factors which have already been mentioned. The salts alone have not measurably affected the interior surface.

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RELATION OF THE MECHANICAL ANALYSIS TO THE MOISTURE EQUIVALENT OF SOILS

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The soil physicist has been very much interested during the past decade in trying to find some constant that will be a measure of the physical properties of soils so that when comparing two or more soils, this so-called constant will express numerically the difference that may exist between such soils.

Comparatively little work has been done by use of the moisture-equivalent determination and since some writers have claimed that there is a direct relation between the mechanical analysis of a soil, for instance, and its moisture equivalent, it was thought desirable to present a preliminary report at this time showing some results that have been obtained in the laboratory of the division of soil technology of the University of California, and to endeavor to offer explanations of the results.

The moisture equivalent was determined by use of the centrifuge designed by Briggs, et al, (2, 3) and was operated at a speed of 2400 revolutions per minute for a period of 30 minutes. This machine exerts a force 1000 times that of gravity on the saturated soil contained in the centrifuge cups.

The mechanical analysis was made by the Bureau of Soils method (4, 6) and special care was exercised to insure that the separation into the seven groups was made as accurately as possible. The analysis was made on 12 different soil types in duplicate, ranging in texture from coarse sand to clay and in origin from residual to recent alluvial. All of the particles belonging to the same group were combined so that a composite sample for each of these seven groups would be obtained. The moisture equivalent for these composite groups of soil particles was then determined in duplicate and not calculated indirectly, as most previous investigators have done.

The results obtained from the moisture-equivalent determinations of these seven grades of soil particles are given in table 1.

Specific-gravity determinations were also made on these seven grades. The results obtained are given in table 2 and show a range from 2.64 to 2.69 and seem to indicate that the separation by the mechanical analysis gave seven grades of soil material that differed mainly in the size of the particles constituting any one group, and not in any marked variation in mineral content.

By noting the results obtained for the different-sized particles making up a soil, as given in table 1, it can readily be seen that textural grade has a definite influence on the moisture equivalent of the soils and that each grade has a definite capacity for holding moisture.

There are soils in the western part of the United States which contain a large proportion of particles ranging from 0.05 mm. to 2 mm. in size, with a relatively small amount of silt and clay. They are, however, capable of holding sufficient moisture for the successful maturity of crops such as grapes, in regions where the annual rainfall averages below 15 inches and no irrigation is resorted to.

TABLE 1
Results obtained in moisture-equivalent determinations

TEXTURAL GRADE	SIZE OF PARTICLES	MOISTURE EQUIVALENT
	mm.	per cent
Fine gravel.....	2.0 -1.0	1.18
Coarse sand.....	1.0 -0.5	1.44
Medium sand.....	0.50 -0.25	1.85
Fine sand.....	0.25 -0.10	2.34
Very fine sand.....	0.10 -0.05	4.62
Silt.....	0.05 -0.005	24.99
Clay.....	0.005-0.0001	61.03

TABLE 2
Results obtained in specific gravity determinations

TEXTURAL GRADE	SPECIFIC GRAVITY
Fine gravel.....	2.67
Coarse sand.....	2.64
Medium sand.....	2.64
Fine sand.....	2.69
Very fine sand.....	2.66
Silt.....	2.65
Clay.....	2.66

The above results indicate that if it is possible to use the mechanical analysis as an indirect method for the calculation of the moisture equivalent, the investigator must give to each textural grade a definite and distinct value and not disregard the sands, or group three or four grades into one.

Three synthetic soils were then made up from these grades of soil particles, following the average mechanical analysis for these three soils as given by the United States Bureau of Soils (8), and the moisture equivalent was determined for these synthetic soils by the usual method. Before determining the moisture equivalent, however, the soils were allowed to stand in a moistened condition for ten days to allow a thorough saturation and an adjustment in the structure of the soil mass. The mechanical analysis and the moisture equivalent of these synthetic soils are shown in table 3.

Various formulas have been proposed for the calculation of the moisture equivalent from the mechanical analysis, of which the following two have received the most prominence:

Briggs (5) proposed the values

$0.02 \text{ sands} + 0.22 \text{ silt} + 1.05 \text{ clay} = \text{moisture equivalent}$,
and Alway and Russell (1)

$0.14 \text{ sands} + 0.27 \text{ silt} + 0.53 \text{ clay} = \text{moisture equivalent}$.

The results obtained up to this time in our laboratory agree more closely with the latter formula than with the former. By noting the results given in table 1 it will be found that the sands total 0.11, the silt 0.25 and the clay 0.61.

TABLE 3
Mechanical analysis and moisture equivalent of three synthetic soils

SOIL TYPE	FINE GRAVEL	COARSE SAND	MEDIUM SAND	FINE SAND	VERY FINE SAND	SILT	CLAY	MOISTURE EQUIVALENT
	per cent	per cent	per cent	per cent	per cent	per cent	per cent	per cent
Loam soil.....	2	5	5	15	17	40	16	21.02
Sandy loam.....	4	13	12	25	13	21	12	14.50
Fine sandy soil.....	1	4	10	57	17	7	4	6.65

TABLE 4
Determined moisture equivalent compared with calculated moisture equivalent

SOIL TYPE	MOISTURE EQUIVALENT		
	Determined	Calculated	Departure
Loam soil.....	21.02	20.86	-0.16
Sandy loam soil.....	14.50	14.01	-0.49
Fine sandy soil.....	6.65	6.32	-0.33

Taking the values for the moisture equivalent as determined for each separate and then calculating the moisture equivalent by using these values in conjunction with the mechanical analysis of the synthetic soils and comparing the result with the actual moisture equivalent of the synthetic soil, the value of a formula can be tested. The averages used for the various textural grades on the basis of the results given in table 1 were: fine gravel 0.010, coarse sand 0.015, medium sand 0.020, fine sand 0.020, very fine sand 0.045, silt 0.250, and clay 0.600.

The results of the determined moisture equivalent compared with the calculated moisture equivalent for the three synthetic soils are given in table 4.

If the sands are totaled the departure of the calculated moisture equivalent from the determined moisture equivalent is much greater than if definite values are given to each of the sand groups, as was done in the table.

The results given in table 4 show that the calculated moisture equivalent

is practically the same as the determined when separate values are given to the seven individual grades of texture, and not when determined by totaling the five grades of sand or disregarding the sands and just considering the silt and clay content of a soil.

A mechanical analysis was then made on 18 samples of soil, the moisture equivalent determined by use of the centrifuge as well as calculated, and the departure noted. The calculated moisture equivalents were in nine cases below the determined and in the others above. The departure of the calculated moisture equivalent from the determined ranged from -0.90 to +2.94.

On a similar set of samples (ten in number) the mechanical analysis and the moisture equivalents were determined by some advanced students in our laboratory. The departure of the calculated moisture equivalent from the determined was greater than that found in the previous set and ranged from -5.44 to + 5.31.

For the third set of samples the mechanical analysis for 30 samples of soil were obtained from the report of the Bureau of Soils, The Soil Survey of the Ukiah Area, California. Duplicate samples of each of these soils were in our laboratory and the moisture equivalent was determined on them. The calculated moisture equivalent departed from the determined from -0.57 to + 7.28.

There were several samples of special interest in that the mechanical analysis was practically the same, yet the determined moisture equivalent varied considerably. The results for some of these peculiar samples are given in table 5.

The mechanical analyses of the first two samples given in table 5 are almost identical, but the determined moisture equivalent of one is 30.80 and for the other 35.82. A similar condition exists for the other two samples noted in the same table.

Taking the average mechanical analysis of several representative soil classes as given by the Bureau of Soils (8) and calculating their moisture equivalent by using the values for the various textural grades as given in table 1, the results shown in table 6 were obtained.

Table 6 shows that from the average of a large number of mechanical analyses there is a direct relation between texture of the soil and its moisture equivalent.

Alway and Russell state (1) "If the mechanical analysis is to be used for the indirect determination of the moisture equivalent, it will be necessary at least in the case of some widely-differing soil types to employ several different formulas."

From the results given in this and previous publications it becomes evident that one formula will not hold in all cases, if that formula is calculated by means of least squares as was done by Briggs and McLane (2, 3) or by direct determination of the moisture equivalent for the various separates, as was tried in this laboratory.

One factor that has been overlooked by most investigators has been the influence of the shape of the soil particles on the moisture retentiveness of soils or on their moisture equivalent. Free states (7), "A very micaceous soil, for instance, has very different physical properties from one of the same mechanical analysis but composed of particles which are mainly spheres instead of disks." The results reported in this paper for the three synthetic soils show that the calculated moisture equivalent agrees very closely with the determined moisture equivalent, keeping in mind that the same sepa-

TABLE 5
Mechanical analysis and moisture equivalent of four special samples of soils

DESCRIPTION OF SAMPLE	MECHANICAL ANALYSIS							MOIS-TURE EQUIV-ALENT DETER-MINED
	Fine gravel	Coarse sand	Medium sand	Fine sand	Very fine sand	Silt	Clay	
	per cent	per cent	per cent	per cent	per cent	per cent	per cent	per cent
Residual surface soil (Climax clay adobe).....	2	2	1	6	9	33	47	30.80
Residual subsoil (Climax clay adobe).....	1	3	1	6	9	34	46	35.82
Recent alluvial surface soil (Yolo silty clay loam).....	1	3	2	8	14	47	25	25.97
Recent alluvial subsoil (Yolo silty clay loam).....	2	3	2	11	14	45	23	21.33

TABLE 6
Calculated moisture equivalents of seven soils

SOIL TYPE	MOISTURE EQUIVALENT CALCULATED
Coarse sand.....	6.38
Sandy loam.....	14.00
Fine sandy loam.....	15.04
Loam.....	20.85
Silt loam.....	25.91
Clay loam.....	26.11
Clay.....	34.82

rates were used for the derivation of the formula as were used in making the synthetic soils. When one, however, applies the results obtained to field soils varying widely in age and origin, the results are far from being in agreement.

It was thought at first by the writer that it would be possible to have one formula to be used for residual soils, another for "wind-laid," another for recent alluvial, etc.; which might take care of the shape of the soil particles. When one, however, sees how the surface soil of residual origin as given in table 5 has the same mechanical analysis as the subsoil of the same origin

and yet a considerably lower moisture equivalent, while on the other hand, a recent alluvial surface soil has the same mechanical analysis as its subsoil yet a considerably higher moisture equivalent, it is evident that any suggested formulas for calculating a constant such as the moisture equivalent from the mechanical analysis of soils are far from accurate.

The mechanical analysis just considers the amounts of the various-sized particles in a given soil, while the moisture equivalent as determined by means of the centrifuge gives a soil constant which is influenced not only by the size and amount of the different particles present in a soil, but also by the shape of the soil particles, amount of organic matter present, amount of soil colloids present, chemical composition of the soil, etc.

From the data given it is felt that while the moisture equivalent calculated from the mechanical analysis according to the formulas suggested gives approximate results, nevertheless they are far from accurate for scientific work, and it will be necessary to make an actual moisture-equivalent determination for satisfactory results.

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THE VOLUMETRIC DETERMINATION OF SULFATES IN WATER EXTRACTS OF SOILS

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The investigation of the water-soluble plant-food elements in soils offers one of the most fruitful methods of attacking problems in soil fertility. Of these elements, considerable interest has been evinced of late in the sulfur requirements of crops and the use of sulfur fertilizers. A number of recent articles on this subject suggest that possibly the sulfur nutrition of plants has not received sufficient attention. During recent investigations in this laboratory as reported by Stewart (4), it became desirable to ascertain the exact amounts of sulfates present in water extracts of cropped and uncropped soils at all seasons of the year.

For a correct determination of the elements extracted from soils by water, the usual gravimetric or volumetric methods do not suffice, owing to the minute quantities present. The first compilation of special methods used in the analysis of water extracts of soils was presented by the Bureau of Soils (3) in 1906. Of these methods, the turbidity method for sulfates as described by the authors, Schreiner and Failyer, was first used. The determination of sulfates by this method consists in comparing the turbidity produced by the addition of barium chloride to the water extract with that produced in a standard sulfate solution treated similarly. After a thorough trial this method was discarded by us, inasmuch as concordant results were not obtained. When from 10 to 20 parts per million of sulfate were present, fairly accurate results were obtained but with less than 10 parts per million the results were generally 30 to 40 per cent in error.

The colorimetric method for sulfate as described by Winkler (5) and quoted by the Bureau of Soils (3) also was investigated. This method consists in precipitating the sulfate as barium sulfate by means of barium chromate in acid solution and upon neutralizing, an amount of chromate equivalent to the precipitated sulfate remains in solution. This is estimated by comparison with a standard chromate solution. Owing to the fact that barium chromate has a solubility of one part in 46,400 parts of water, it was found necessary to correct the final result by subtracting 8.2 parts per million from the number of parts per million of sulfate found. Since nearly all the soils used in this investigation, on being extracted with from 1 to 5 times their weight of water,

yielded solutions containing less than 8 parts per million of SO_4 , the use of this method for small amounts of sulfate was undesirable.

Our attention was called to a volumetric method for the estimation of small amounts of sulfate in urine, as described by Raiziss and Dubin (2). This method depends upon the separation of the sulfate as insoluble benzidine sulfate followed by titration with N/10 KMnO_4 . A recent publication by Drummond (1) advocates titration with N/50 KOH. The permanganate titration, however, seems preferable for very small amounts of precipitate, owing to the fact that a relatively larger amount of permanganate is required for titration (approximately 17 times more N/50 KMnO_4) than N/50 KOH for the same amount of benzidine sulfate. The method of Raiziss and Dubin (2) has been modified by us for the determination of small amounts of sulfates in water extracts of soils. In the investigations referred to above (4), 200 cc. of a 1 to 5 extract, representing 40 gm. soil, has been found in all the 14 soils examined, to contain sufficient sulfate for a convenient determination, using N/20 KMnO_4 . The amount of extract employed should be chosen so as to contain not less than 0.1 mgm. nor more than 5.0 mgm. of SO_4 . The method follows.

Evaporate a suitable aliquot to dryness in a 200 cc. casserole on the steam bath. Ignite at a low heat to destroy organic matter. Take up with about 5 cc. of dilute HCl (1: 9), digesting on a steam bath for 5 to 10 minutes. Filter through a 5.5 cm. filter into a 500-cc. wide-mouthed Erlenmeyer flask, washing with several successive small portions of hot water until the volume amounts to 15 to 20 cc. Cool. Add one drop of phenolphthalein indicator and make just alkaline with NaOH (10 per cent). Acidify with one drop of HCl (1: 4). Add 10 cc. of benzidine hydrochloride (8 gm. per liter of water) and allow to stand 15 minutes with occasional shaking. Filter with gentle suction on a small asbestos pad supported by a porcelain plate in a glass filter tube. Wash with three 5-cc. portions of cold water. Return the asbestos and precipitate to the flask by means of a jet of water and make the volume to about 200 cc. Add 1 cc. of 10 per cent NaOH and boil for 5 minutes. Cool to room temperature, add 20 cc. of concentrated H_2SO_4 and titrate with N/20 KMnO_4 until a pink coloration persisting 20 seconds is obtained. During the titration, the strong yellow color which first appears gradually fades and the solution becomes practically colorless before the end point is reached.

Raiziss and Dubin (2) state that under the above conditions 1 cc. of N/10 KMnO_4 equals 0.099 mgm. of S. By calculation, 1 cc. of N/20 KMnO_4 equals 0.15 mgm. of SO_4 , which is taken as the factor. Next to a careful adjustment of the acidity, the most important precaution has been found to be that of washing. When less than 15 cc. of wash water were used, high results indicated that the excess reagent had not been entirely removed while washing much in excess of 20 cc. caused low results, due to the slight but appreciable solubility of benzidine sulfate.

In order to test the accuracy of the method, sulfate solutions of known con-

centration were analyzed. These solutions had been carefully prepared by dilution of stronger solutions checked by the usual barium chloride method. These results as well as 6 analyses of a typical 1 to 5 soil extract are given in table 1. The method is believed by us to be more dependable than the turbidity or colorimetric methods previously described, especially for relatively small amounts of sulfate.

TABLE 1
Determination of sulfate by volumetric method

	ANALYSES OF KNOWN SOLUTIONS						ANALYSES OF A SOIL EXTRACT	
	N/20 KMnO ₄	SO ₄	N/20 KMnO ₄	SO ₄	N/20 KMnO ₄	SO ₄	N/20 KMnO ₄	SO ₄
	cc.	p.p.m.	cc.	p.p.m.	cc.	p.p.m.	cc.	p.p.m.
Theoretical.....	1.0	0.75	10.0	7.50	20.0	15.00		
Found.....	0.8	0.60	9.5	7.12	19.6	14.70	4.9	3.67
Found.....	0.9	0.68	9.6	7.20	20.0	15.00	4.8	3.60
Found.....	1.1	0.83	9.6	7.20	19.9	14.92	4.8	3.60
Found.....	0.9	0.68	9.8	7.35	19.8	14.85	4.7	3.53
Found.....	1.1	0.83	9.7	7.27	19.9	14.92	4.8	3.60
Found.....	1.0	0.75	9.7	7.27	20.5	15.37	4.7	3.53
Average.....	0.97	0.73	9.65	7.23	19.95	14.96		
Maximum error.....		20 per cent		5.0 per cent		2.5 per cent		
Average error.....		3 per cent		3.5 per cent		0.25 per cent		

SUMMARY

The volumetric determination of small amounts of water-soluble sulfates in soils by titration of the precipitated benzidine sulfate with potassium permanganate is described.

The method is shown to have an average error of 3 per cent and is believed to be superior to colorimetric or nephelometric methods, especially for small amounts of sulfate.

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A STUDY OF THE ROOT-NEMATODE (HETERODERA RADICICOLA) AND ITS CONTROL

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INTRODUCTION

The root-nematode, eel-worm or root-gallworm, has long been known in the United States, particularly in soils of the South and occasionally in soils of the northern states. It is also widely distributed in greenhouses and the direct or indirect damage done by the nematode is very great. Up to date, an entirely satisfactory remedy has not been found, consequently this aspect of the problem offers a rich field for investigation. At the outset, then, it was necessary to consider all possible phases, and as a result certain points are revealed in this study which are of general interest, but these do not directly affect or concern the problem of practical control.

Since the discovery of the root-nematode in 1855 by Rev. M. J. Berkeley, it has been reported as occurring on 480 different species of plants, some of which are the tomato, celery, beet, lettuce, lima bean, radish, spinach, egg-plant, gardenia, carnation, sweet-pea, rose, violet, ginseng and other plants which are not so common.

Nematode infestation becomes noticeable when the plants show dwarfing and have a general unhealthy appearance. In some cases, however, infestation is not apparent until the roots are examined. An infested root is usually enlarged at the tip as a result of the irritation produced by the nematode. In this respect it differs from the round and lateral tubercles of bacteria formation, and furthermore, the swellings due to the root-nematode are elongated and pear-shaped.

The larva, or early stage in the animal's development, is a slender transparent eel-like organism which moves about by a whipping process. In a liquid containing no sediment this thrashing about does not produce a change of location while in a solution containing small particles the thrashing results in locomotion due to friction against the particles. The larva is about 400μ or approximately 0.5 mm. in length and nearly 15μ in width. It is colorless, but the body-wall and particles of food within the animal serve to differentiate it from the medium in which it is located. The larva, on hatching from the egg, searches for a growing root and upon finding it, enters at the growing tip and a gall is subsequently formed. Many larvæ may enter a single root but

the size of the gall and the number of larvæ within are not necessarily correlated.

While in the root, the larvæ undergo several molts and become true males and females. The mature male is similar to the larva in form and appearance. It is, however, much larger, being about 1400μ in length and 30μ in thickness (fig. 1). The female becomes pear-shaped and is from 400 to 1000μ in length, of a pearly white color. After being fertilized by the male the eggs begin to form.

Brown, oval eggs are forced out by the female. These eggs measure about 90μ in length and 40μ in thickness. Repeatedly, larvæ were observed coiled within the thick shell of the egg. (Fig. 1.)

Under average greenhouse temperatures, the eggs hatch in 5 days and the newly-hatched larvæ may immediately attack susceptible plants. The entire life-cycle in a greenhouse is completed in about 30 days.

The larvæ are capable of encysting and thereby capable of resisting adverse conditions, such as drought and certain chemicals. The eggs are likewise very resistant and this stage is able to withstand many strong chemicals.

Before 1898 (10) very little attention had been given to the study of nematode control; the limited amount of investigation before this period dealt mainly with the morphology and physiology (1). From the standpoint of control, steam sterilization and some chemicals have been tested (2, 3, 7, 8). Satisfactory control is claimed by steam sterilization of the soil but this method is in many cases impossible or at least impracticable. A large number of chemicals (8, 10) has been tried but so far as known none has given satisfactory results. In view of the above facts, it was thought necessary to consider all phases of this problem that time would permit.

EXPERIMENTS

Moisture

The first experiment tried was to determine the effect of moisture on root-nematode activity.

In the preliminary experiments with moisture it was seen that galls, taken from infested gardenia plants and placed in water over a source of mild heat, produced very active nematodes. For further study galls of approximately the same size (3.5 x 3 mm.) were thoroughly washed in distilled water and placed singly in slender dishes and each was treated, in a duplicate series, with different quantities of distilled water and maintained at a constant temperature for 72 hours and examined under the low power of a microscope at different intervals of time. All observations were from 1 hour to 72 hours. The results were the same at the end of 1 hour as at the end of 72 hours. The observations are given in table 1. In the table, Series A and B are duplicates and have the same treatment.

Another moisture experiment was made to confirm the preliminary one. This was run in triplicate series, each series having the same treatment. Results are given in table 2.

It can be seen from table 2 that the activity of the root-nematode is increased, the temperature remaining constant, with an increase in moisture

TABLE 1
Effect of moisture on root-nematode activity (at 21°C.).

WATER cc.	SERIES A	SERIES B
0.0	Inactive	Inactive
0.1	None out	None out
0.2	Very few out and slightly active	None out
0.3	Very few out and active	Few out and slightly active
0.4	Very few out and active	Few out and slightly active
0.5	Many out and active	Many out and very active
0.6	Many out and not active	Many out and few active
0.7	Immerged, few slightly active	Immerged but inactive
0.8	Immerged inactive	None seen
0.9	None seen	None seen
1.0	None seen	None seen
2.0	None seen	None seen
5.0	None seen	None seen
10.0	None seen	None seen

TABLE 2
Effect of moisture on root-nematode activity (at 19°C.).

WATER cc.	SERIES A—LARVAE	SERIES B—LARVAE	SERIES C—LARVAE
0.0	Inactive	Inactive	Inactive
0.2	One out and active	One out and active	None seen
0.3	Slightly active	Slightly active	Slightly active
0.4	Some very active	Slightly active	Some very active
0.5	Many very active	Many active	Some very active
0.6	Many very active	Many active	Some very active
0.7	Many very active	Many active	Slightly active
0.8	Few active	Slightly active	None seen
0.9	None seen	Slightly active	None seen
1.0	Slightly active	Slightly active	Inactive

until an optimum moisture content is reached, and when continued above that point the activity is decreased.

In the above experiments, sterile water was used in order to eliminate the toxic substances, such as chlorine, copper salt, etc., found in tap water. This introduced another factor—that of plasmolysis. It was suggested that the concentration of sterile water was below the concentration of the liquid in the

body of the nematode and therefore plasmolysis might occur. This theory was tested by placing gardenia galls in a series of stender dishes containing sterile water and a similar series using water extracted from the soil—the natural solution in which the nematode normally lives. It was observed that the results from both solutions (0.2 to 1.0 cc.) compared very closely.

This proved that sterile water had no plasmolyzing effect used in rates up to 1 cc., and that the experiments were therefore sufficiently accurate.

Also, in connection with this experiment it was thought that because a definite number of larvæ were not used in each stender dish results would not be accurate. This possibility of error was dismissed on the ground that sufficient numbers (200 to 400) of larvæ were present in each dish to give safe averages. In every test the gall was crushed in order to reveal the dormant larvæ in observations recorded as "none seen." The fact that some larvæ emerged and others did not may be due to their location in the gall, that is, some of them were not near enough to the surface of the root to be affected by the moisture. The galls became submerged with 0.7 cc. of water.

If in the above tables on moisture, "inactivity" means death, then flooding the soil might offer a remedy for nematode infestation. Furthermore, it has previously been observed in the South that the root-nematode does not occur in bogs or water-soaked lands. Many observers have noticed that the nematode occurs mostly in sandy soils and not in clays. This may be due to the fact that sand does not retain water for any considerable length of time and thus allows sufficient aeration for the nematode. On the other hand, clay soils retain moisture and thus reduce the amount of air held. For the majority of cases, therefore, flooding was considered impracticable, and an unsatisfactory control, except perhaps under special conditions, and was dismissed from further consideration.

Temperature

The next test made was to determine the effect of temperature on nematode activity.

As in the experiment on moisture, gardenia galls of approximately the same size (3 mm. x 3.5 mm.) were immersed in 0.4 to 1.0 cc. of distilled water and triplicate series placed under three different temperatures. To keep the moisture constant the stender dishes were placed in tin trays containing water, and were covered with cheese-cloth which rested in the water and was kept moist by capillary attraction.

Series I was placed out-of-doors, sheltered from rain. The temperature averaged about 8.8°C., or 48°F.

Series II was placed in an electric incubator at a temperature from 36.6° to 38.8°C., or 98° to 102°F.

Series III was placed in a greenhouse at a temperature of 26.6°C., or 80°F. The contents of all dishes were examined at the end of 24 hours under the

low power of a microscope. The results are given in table 3. Here again Series A, B and C have the same treatment.

In the above table "none out" and "none seen" indicate that no larvae had emerged, but at the end of this experiment each gall was crushed and inactive larvae were found.

The observations recorded in table 3 seem to warrant the conclusion that the optimum temperature for the nematode is between 65° and 85°F. Generally speaking, temperatures above 65°F. increase nematode activity, and temperatures below 65°F. and above 85°F. decrease nematode activity. The animal may be killed by freezing (32°F.), and by heating to 101°F. the larvae and eggs are killed.

TABLE 3
Effect of temperature on root-nematode activity

	WATER α.	I (AT 8.8°C.)	II (AT 36.6° TO 38.8°C.)	III (AT 26.6°C.)
Series A . . .	0.4	Out and active	Out and inactive	Out and inactive
	0.6	Out and active	Out and inactive	None out
	0.8	None out	Out and inactive	Out and slightly active
	1.0	Out and active	Out and slightly active	Out and slightly active
Series B . . .	0.4	Out and active	Out and inactive	Out and very active
	0.6	Out and active	Out and inactive	Out and very active
	0.8	Out and active	Out and inactive	Out and very active
	1.0	Out and active	None out	Out and very active
Series C . . .	0.4	Out and active	None out	Out and very active
	0.6	Out and active	Out and active	Out and very active
	0.8	Out and active	Out and inactive	Out and very active
	1.0	Out and active	None seen	Out and very active

The above results suggested that raising the temperature of the greenhouse soil to 101°F. would kill the larvae and eggs. This however did not appear to be practicable for most greenhouses. High temperature may be applied with some degree of success at least, by exposing infested soil to the hot rays of the sun and turning it over to insure thorough sterilization.

Chemicals

Failing to discover a satisfactory remedy for nematode attack in the study of moisture and temperature conditions, attention was next directed to the study of chemicals. At the outset it was seen that a great many of the favorite chemicals used in soil sterilization could be omitted, for J. A. McClintock (8) has shown that the nematode could not be satisfactorily combated by any of the following: carbon disulfide, corrosive sublimate, calcium carbide, ammo-

nium sulfate, formalin, nicotine, benzine or kerosene. Furthermore, Stone and Smith, in Massachusetts (10), demonstrated that the following chemicals are ineffective: manganese sulfate, common salt, potassium nitrate, magnesium sulfate, calcium sulfate, kainit, sodium nitrate and lime.

The recent success with sodium cyanide (NaCN) obtained by Dr. T. J. Headlee of the New Jersey Agricultural Experiment Station, as a soil sterilizer in the control of wire-worms suggested that the same treatment might be used in combating the root-nematode.

Gardenia galls were placed in stender dishes as in experiments with moisture and temperature and treated with amounts of cyanide, ranging from 0.7071 gm. to 0.0707 gm. dissolved in 10 cc. of water, giving solutions from 0.7 to 7 per cent concentration. On standing 48 hours the larvæ had completely disappeared. This disappearance may have been due to disintegration. The eggs also were attacked and made colorless.

The above experiment was repeated in duplicate, with 2 mg. NaCN dissolved in 0.5 cc. of water (shown by the first experiment to be the approximate optimum moisture). The results are given in table 4.

TABLE 4
Sodium cyanide treatment (at 18°C.)

	A—LARVÆ	B—LARVÆ
1	None out	Dead
2	Dead	Dead
3	Dead	Dead
Check (no NaCN)	Very active	Slightly active

On crushing the gall in the cylinder 1A, the larvæ were found to be dead. This test shows conclusively the high killing effect of cyanide on the larvæ. This strength of cyanide (25 per cent) did not affect the eggs.

Sphagnum Moss

It has been noticed that roots of plants growing in sphagnum moss did not become infested with nematodes while roots on the plants not in sphagnum moss became infested.

This interesting point was summarily tested. Dry sphagnum moss was placed in a beaker of distilled water and boiled for 10 minutes. The mixture was then filtered and the moss extract applied at the rate of 0.5 cc. to gardenia galls in stender dishes as in previous experiments. The observations showed that the moss extract killed the larvæ (table 5).

TABLE 5
Effect of sphagnum moss extract treatment (at 18°C.)

	cc.	A	B
1	0.5	Dead	Dead
2	0.5	Dead	Dead
3	0.5	Dead	Dead
Check		Very active	Very active

Formaldehyde

A similar experiment was also tried with 40 per cent formaldehyde and the results are given in table 6.

This demonstrates the high killing effect of formaldehyde.

TABLE 6
Effects of formaldehyde (40 per cent) treatment (at 18°C.)

	cc.	A—LARVÆ	B—LARVÆ
1	0.5	Dead	Dead
2	0.5	Dead	Dead
3	0.5	Dead	Dead
Check		Very active	Very active

DISCUSSION OF RESULTS

The root-nematode (*Heterodera radicicola*) has been a serious pest to plants growing in certain soils and particularly in greenhouse soils. The investigations concerning this pest have been mainly morphological and the attempts to control it have thus far not given entirely satisfactory results. Steam sterilization of the soil does give complete control where it can be practically applied and with thoroughness, but the average conditions do not offer a way for the application. For example, greenhouses fitted for hot-water heating are not adapted to the use of steam. It was for such a condition that the attempt was made to find another method.

The first experiment was to determine the effect of moisture upon the nematode and it was observed that moisture is a necessity for activity but if the water content is in excess, then the animal becomes inactive. So there is an optimum moisture for the nematode.

An experiment to test the effect of temperature demonstrated that the nematode is inactive at comparatively low temperatures and at high temperatures, and its greatest activity is at a temperature from 65° to 85° F. So there is also an optimum temperature.

The recent success with sodium cyanide suggested this treatment as a possible remedy and it was found to be very effective in killing the larvæ and in a moderately concentrated solution to destroy the egg stage.

Similarly sphagnum moss extract and formaldehyde were found to be toxic to the nematode.

GREENHOUSE EXPERIMENTS

The satisfactory results of different treatments in the laboratory led to experiments with soil in the greenhouse. Fortunately, a very thoroughly infested greenhouse bench offered itself for the experiment. This bench had just produced tomato plants which were thoroughly infested with nematodes.

The tomato plants were removed and the soil raked clean of the roots and other objectionable materials and turned over thoroughly. Next the bench was divided into 7 plots, each separated by 5 inches of air space to prevent migration of nematodes from one plot to another.

Plots 2, 4 and 7 were used as check plots. Plots 1 and 3 were treated with sodium cyanide (NaCN) at the rates of 100 pounds and 200 pounds per acre (or 0.036 ounce and 0.072 ounce, respectively). The cyanide was dissolved in water at the rate of 1 gallon of water to $\frac{1}{16}$ ounce and $\frac{1}{8}$ ounce of cyanide, respectively.

The solution was applied to the soil with a sprinkling can at the rate of $\frac{1}{2}$ gallon per square foot of soil, and at the same time the soil was turned back and then thrown over to cover the solution. In this way every particle of soil received the same strength of the solution. After 4 days, the soil was broken up and aerated.

Another similar application was made to these two plots 5 days after the first application. This was done on the theory that the eggs of the animal might have resisted the first application and the larvæ would have hatched at the end of 5 days under the greenhouse temperature (70° to 80° F.) and would be killed by the second application of cyanide.

At the end of 3 days these soils were turned over and completely aerated to remove all traces of cyanide gas. The soil in these and check plots was all the time kept moist and in good tilth. At the end of another 3 days (February 7, 1917) these two plots and the check plots, No. 2 and 4, were planted to cucumbers, radishes and tomatoes. Mice attacked the seeds and necessitated replanting the cucumbers and radish seeds.

At this time it was thought that the mice might have carried some particles of soil from the infested check plots, so note was made of this partly to explain any error that might occur.

Plots 5 and 6 were treated with sodium cyanide at the rates of 50 and 100 pounds, respectively, to ascertain whether a weaker application would control the nematode, as laboratory experiments seemed to show, and whether or not one treatment would be sufficient. The same method of cyanide application was used as with plots 1 and 3. Plot 7 was reserved as a check plot. These plots were aerated 3 days after and planted 6 days after the application to lettuce, radishes, and tomatoes.

The percentage of seed germination was good and the seedlings grew well for about 3 weeks, when it became evident that they were not making average growth. Then it was realized that the soil was low in available nitrogen and tankage was subsequently applied at the rate of 10 gm. per square foot (or approximately 800 pounds per acre). In a few days the plants began to show the effect and made a rapid growth.

While waiting for the plants in these plots to make sufficient growth to observe results, several other experiments were performed.

The question of the effect of the application of sodium cyanide on plants growing in soil was tested by potting up 4 infested tomato plants in infested soil and allowing them to grow under normal conditions for 2 weeks. One of these plants was treated with cyanide dissolved in water and applied at the rate of 50 pounds per acre and another at 100 pounds per acre, which were amounts found to kill the nematode in the laboratory. The remaining two plots were used as checks. The plants in the treated pots wilted at the crown at the end of one day, and at the end of 5 days were completely dead, while the check plants grew as normally. Galls were examined from all plants and the nematode larvæ in them were found to be alive and very active.

The results of the greenhouse experiments were recorded from the time the plants had made a growth of 6 inches up to the time when they were 10 inches high (April 21, 1917). The plants were 18 weeks old at the time the final observations were recorded (table 7).

TABLE 7
Greenhouse experiments with cyanide treatment

DATE	NUMBER OF PLANTS EXAMINED	Plot 1			Plot 2			Plot 3			Plot 4			Plot 5			Plot 6			Plot 7		
		Galls			Galls			Galls			Galls			Galls			Galls			Galls		
		S*	M	L	S	M	L	S	M	L	S	M	L	S	M	L	S	M	L	S	M	L
March 31, 1917....	20	56	9	0	148	10	0	0	0	0	101	15	0	8	3	0	17	0	0	125	7	0
April 11, 1917....	18	30	9	1	130	9	7	2	0	0	40	0	0	15	0	0	60	2	0	37	0	0
April 18, 1917....	15	35	7	0	100	3	4	1	0	0	36	0	0	19	0	0	130	3	0	110	0	0
April 21, 1917....	27	30	3	10	120	7	13	0	0	0	57	2	2	30	0	0	110	3	2	130	10	15

*S = small M = medium L = large

This experiment demonstrated that cyanide to be effective must be applied in larger quantities than 100 pounds per acre to kill the larvæ, and under such applications the soil must be free from growing plants. As shown by the previous experiment, seed will germinate and plants will grow well in soil which has been treated with sodium cyanide used as high as 200 pounds per acre in two applications, and which has been well aerated after the application.

The table of observations shows that the treatment of 200 pounds of cyanide per acre followed by a second treatment 5 days later gives nearly a perfect control. Out of 80 plants examined only 3 galls were found. This is plainly

within experimental error and the results warrant the recommendation of this method of sterilization.

Due to the fact that the treated and untreated plots were so close together, some of the particles of soil and water carrying the nematodes could easily have been transmitted from one plot to the other.

There was no apparent difference in growth of plants in the various plots. They all were very healthy-looking at 18 weeks after the seed was planted. The experiment was then discontinued.

SUMMARY

1. A serious pest to plants, growing in warm soils and especially in greenhouse soils, is the root-nematode (*Heterodera radicicola*) sometimes called the eel-worm or root-gallworm.

2. This parasite is a minute organism which penetrates the roots of plants causing them to become enlarged and deformed, and resulting in a hindrance to growth.

3. The nematodes may be distributed by their own motion, by water, in soil clinging to implements, by infested plants transferred to an uninfested soil, by seed, by cuttings, and by manure.

4. The following conditions are necessary for their growth and reproduction: (a) an optimum moisture, (b) an optimum temperature, (c) food, and (d) oxygen.

5. The following were found to decrease nematode activity and may be applied as methods of control: (a) excess of moisture, (b) high temperature (101°F.), (c) formaldehyde, (d) sphagnum moss extract, and (e) sodium cyanide (NaCN) dissolved in water.

6. In experiments with greenhouse soils the following treatment gave a satisfactory and practical remedy for nematode infestation:

Sodium cyanide dissolved in water is applied in the proportion of 200 pounds per acre, one-third gallon per square foot of soil. One week after the first treatment a second similar treatment is given. This method is for soils free from plants. The soil should be aerated and leached well before seed or plants are put into it in order to remove the greater amount of cyanide gas.

7. The cost of materials for this treatment is \$112 per acre. For greenhouse soils where steam sterilization is impracticable this is a remedy which is to be recommended.

CONCLUSIONS

The object of our study is thus accomplished. A satisfactory and practical control for the soil nematode is found in sodium cyanide.

In treating nematode-infested soil, the number of square feet of soil should first be determined. The number of pounds of cyanide should then be calculated on the basis of 200 pounds per acre. Next, the amount of water,

figured on the basis of $\frac{1}{2}$ gallon per square foot of soil, should be determined. Dissolve the cyanide in the proper amount of water and apply the solution to the soil with a sprinkler. The soil should be well mixed and kept slightly moist and warm for 5 or 6 days before the treatment in order to bring the larvæ from their cysts and therefore render them more susceptible to the treatment. The third day after the first treatment the soil should be stirred and aerated slightly. The same favorable conditions for nematode activity should be maintained until after the second treatment. About one week after the first treatment the second application determined in the same manner should be given.

Of course, all plants should be removed from the soil before the application of cyanide, for most plants succumb to such a treatment. Before any seeds are planted or plants set in the sterilized soil, the soil should be thoroughly aerated and leached somewhat to remove the traces of cyanide gas.

Using this method of soil sterilization, the cost of the cyanide used would be 400 pounds at \$.28 per pound, or \$112. For greenhouse soil sterilization, this is an economical and entirely satisfactory investment.

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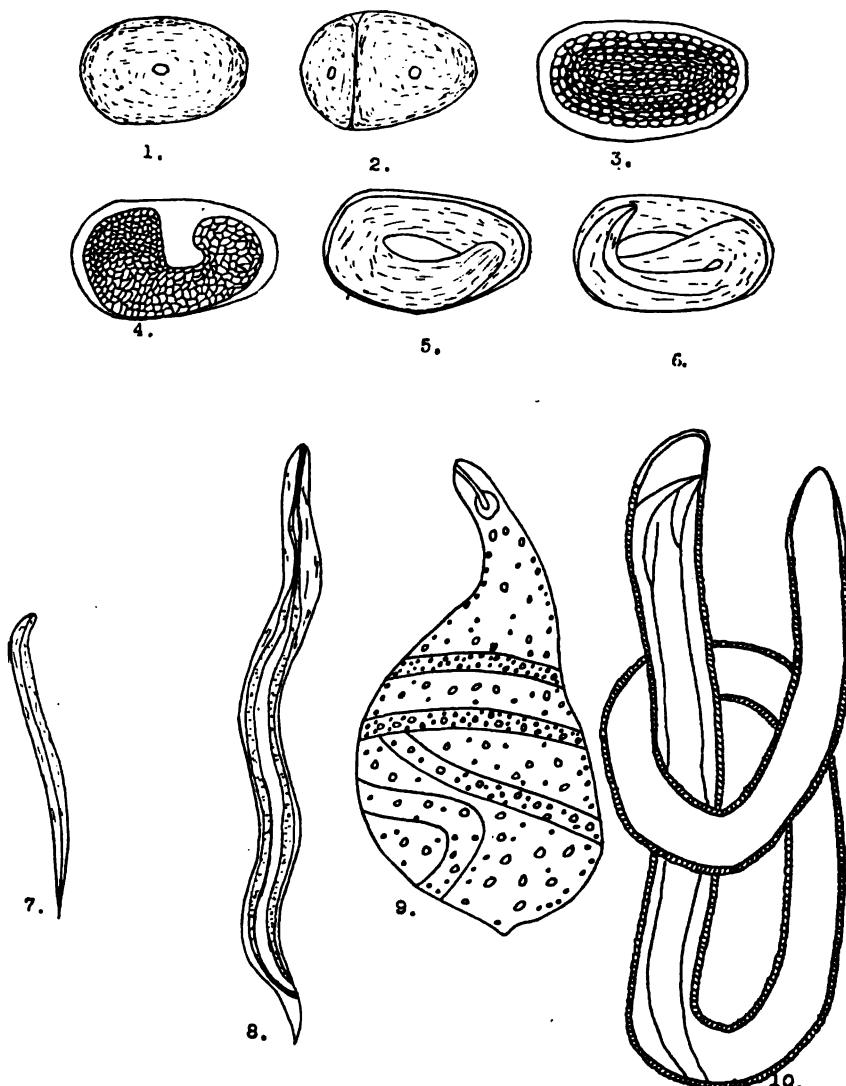


PLATE 1

- Figs. 1 to 6. DEVELOPMENT OF FREE-LIVING NEMATODE
 FIG. 7. YOUNG LARVA
 FIG. 8. IMMATURE FEMALE
 FIG. 9. MATURE FEMALE
 FIG. 10. MATURE MALE

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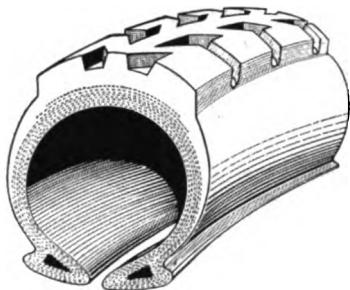
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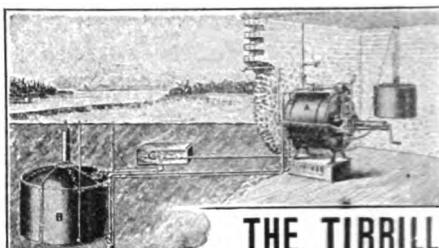
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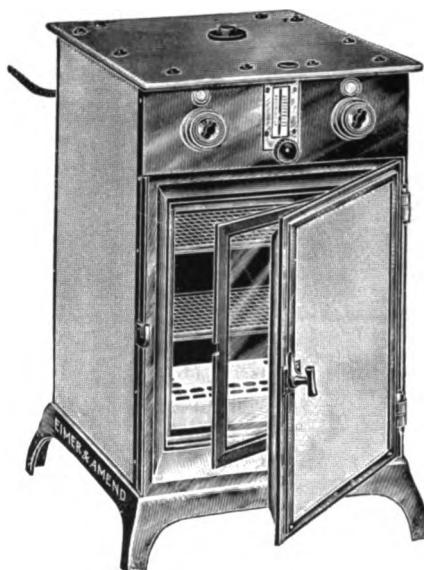
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